by Carnot (10) who showed that many samples of European fossil bone contain much higher amounts of fluorine than bone of modern origin, the fluorine content usually increasing with the age of the bone. This increase, according to Roholm (46), is probably due to a prolonged contact with water or soil containing soluble fluorides. Chemical analysis as well as microscopic and x-ray diffraction examinations indicate that bone is a carbonate-apatite with the probable formula of $3Ca_3(PO_4)_2 \cdot CaCO_3$ or $3Ca_3(PO_4)_2 \cdot CaCO_3 \cdot H_2O$ (6,7,28,29,35). It is logical to believe that the fluorine in water may in some measure replace the carbonate radical in the carbonate apatite, the hydroxyl radical in hydroxy-apatite, the oxygen in oxy-apatite, and the chloride in chlor-apatite with the formation of fluor-apatite, the completeness of the replacement depending on the time of contact and the fineness of subdivision of the "adsorbing" material. Hendricks et. al. (29) state that "upon fossilization, the carbonate group and the water molecule of carbonate apatite are replaced by fluorine". With these assumptions in mind and the knowledge that treated bone would remove fluorine from water, a study was begun of the details of the reaction, the preparation, and the effectiveness of bone in removing fluorine from water. This paper presents the results of such an investigation.

EXPERIMENTAL WORK

Analytical methods used:

The fluorine determination has been a notably weak spot in research on mottled enamel until within the past few years, but now there appear to be several methods available which may be applied to water and solid substances with satisfactory results. When the work on fluorine was begun at the University of Arizona in 1930, only a few methods of quantitatively determining fluorine were available. Results obtained by using these methods were questionable. The volatilization method of Reynolds, Ross, and Jacobs (44), modified for water analysis, was among the first of the methods used by Smith and Smith. The method, however, was long and tedious and was soon abandoned in favor of the Fairchild (23) method as modified and used by Churchill (12). By 1933 several more methods were suggested, and in 1935 H. V. Smith made a comparison of four of the better known and seemingly most reliable procedures for the determination of fluorine (54). The methods studied included those of Willard and Winter (67), Foster (25), Sanchis (49), and the Fairchild (23). Smith found that the first three were in quite close agreement, but that the Fairchild method gave results which averaged two to three times as high as results obtained by the other methods. As a result of this study on the part of Smith, the method of Willard and Winter and that of Sanchis, with slight modifications, were chosen for the work reported in this paper, since they appeared to be the most reliable, the simplest, and the best adapted procedures for the type of work conducted.

The Willard and Winter method (67) consists of the titration of a water soluble fluoride or fluosilicate, in the presence of alcohol, with a standard thorium nitrate solution, using as indicator a lake produced by adding zirconium nitrate to alizarine red. In the presence of fluoride, the lake is decolorized when barely acidified with hydrochloric acid, but regains its pink color when the fluoride has been precipitated as thorium fluoride. The alcohol is used to render the thorium fluoride less soluble. For those cases, however, in which there is present an interfering ion or when the fluorine determination is to be made on a solid substance such as bone or rock phosphate, a steam distillation at 135 degrees centigrade with perchloric acid in the presence of silica is used and the fluorine distilled off as hydrofluosilicic acid. The distillate is then titrated with the standard thorium nitrate. Any ion which forms a precipitate or a non-dissociated salt with fluorine or with thorium, interferes with the titration. Several of such interfering substances are calcium, barium, ferric, aluminum, and phosphate ions.

The mechanism of the reaction is rather simple in theory. The perchloric acid reacts with the fluorine present in the sample to produce hydrofluoric acid, which combines with the silica added to produce silicon-tetra-fluoride. This product in turn reacts with water present to form the hydrofluosilicic acid which is distilled off and titrated with the thorium nitrate. The following series of equations may be given to represent reactions involved:

(1) $2NaF + 2HClO_4 \longrightarrow H_2F_2 + 2NaClO_4$

(2) $2H_2F_2 + SiO_2 \longrightarrow SiF_4 + 2H_2O$

(3) $3SiF_4 + 3H_2O \longrightarrow 2H_2SiF_6 + H_2SiO_3$

The reliability of this method has been questioned from time to time, but, in general, the determination appears to be quite accurate. Considerable controversy has been carried on over the question of loss of fluorine by side-reaction with the silica in the glass of the distilling flask. Instances of etching have been noted after a number of determinations using the same flask. A question has been raised as to the proper distillation temperature; but it now appears to be generally agreed that a temperature of 135 degrees centigrade insures nearly complete recovery of the fluorine. As a result of these controversies, a number of modifications of the original method have been proposed.

The Willard and Winter method for determining fluorine with the use of sodium alizarin sulfonate indicator as proposed by Armstrong (3) has been recently modified as follows: by Hoskins and Ferris (30) who introduced the use of a buffer of monochloroacetic acid; by Armstrong (4) who titrated fluoride in an aqueous rather than in an alcoholic solution, and used silver perchlorate to remove interfering chlorides; and by Churchill, Bridges, and Rowley (13) who eliminated the effect of phosphate by a double distillation. Rowley and Churchill (48) applied the aqueous titration to the determination of quantities of 1 to 50 milligrams of fluorine. Dahle and associates (15) have recently conducted studies on a "back-titration procedure" as well as a number of studies on the temperature of distillation and size of the flask used. Eberz, Lamb, and Lachele (21) studied the titration in alcoholic solution of quantities of 100 to 150 micrograms of fluorine.

In the study presented in this paper, a slight modification of the Willard-Winter procedure was chosen. A double distillation as suggested by Churchill, Rowley, and Bridges (13) was used to further eliminate the effect of the phosphate ion and also to prevent any danger of explosion. Perchloric acid and organic matter form an explosive mixture at high temperatures; hence, a double distillation using sulfuric acid in the first instance and perchloric acid in the second was utilized so as to minimize any danger of explosion and in addition to further separate the fluorine from interfering substances. This double-distillation was found necessary since the organic matter content of some of the bone samples was quite high.

The second modification used was that suggested by Rowley and Churchill (48). These investigators proposed a method whereby the titration of the fluoride can be accomplished in an aqueous solution merely by the addition of a buffer solution prepared by dissolving monochloroacetic acid in a sodium hydroxide solution. The proposed titration procedure appears to be quite accurate, and, since it is somewhat simpler than the originally suggested method, this modification was used in preference.

The Willard and Winter procedure as modified and used in this study follows.

With no interfering elements present:

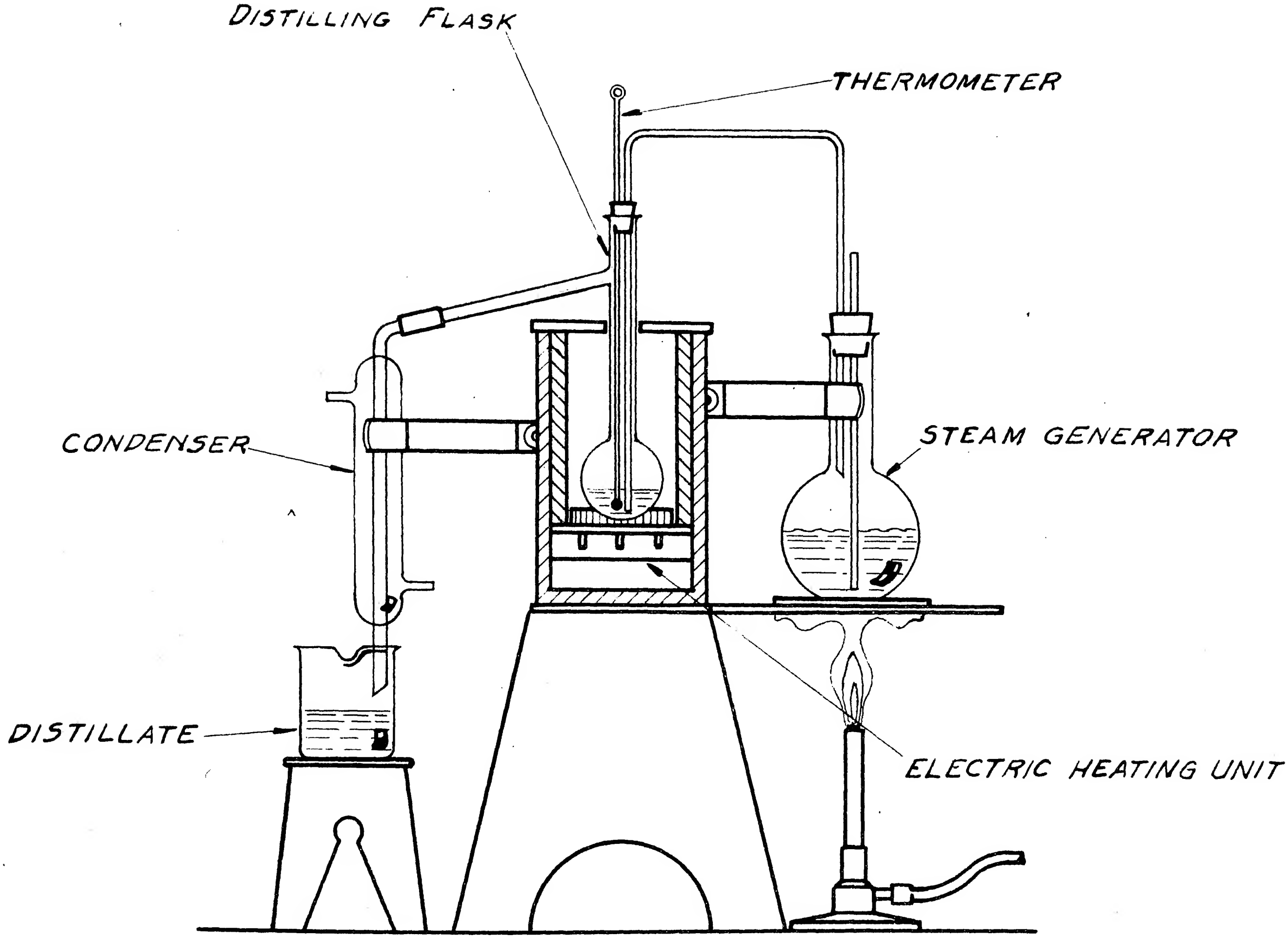
The reagents used for determining fluorine in solution when no interfering elements are present are as follows: (1) Zirconium nitrate, 1 gram of $Zr(NO_3)_4 \cdot 5H_2O$ in 250 cc. of water. (2) Alizarin red, 1 gram of sodium alizarin sulfonate in 100 cc of ethyl alcohol. Filter off the undissolved residue and add 150 cc. of ethyl alcohol to the filtrate. The two solutions are kept in stock and mixed, three parts of (1) and two parts of (2) as needed. Or a 0.05% aqueous solution of sodium alizarin sulfonate may be used. (3) Thorium nitrate solution, standardized against a known fluoride solution. (4) Hydrochloric acid, approximately 1 to 200. (5) Sodium hydroxide solution approximately 2%. (6) Buffer, by dissolving 9.448 grams of monochloroacetic acid and 2.000 grams of sodium hydroxide in 100 cc. of water.

Procedure::

Take an aliquot of 50 to 100 cc. of water sample depending on the amount of fluorine present and the concentration of the titrating solution. Dilute to 100 cc. with water. Add 8 drops of sodium alizarin sulfonate indicator (0.05%). Adjust the acidity with 2% sodium hydroxide and the 1:200 hydrochloric acid solution, finally leaving the solution just acid and the pink color discharged. Add 1 cc. of the buffer solution and titrate with standard thorium nitrate (1 cc. equals 0.25 mg. fluorine) to a permanent pink. Make a blank determination to determine the amount of the thorium nitrate which combines with the indicator.

With interfering substances present:

If the determination is to be made on a water or other liquid, a suitable amount of the solution is reduced on the steam bath to about a 50 cc. volume in the presence of free alkalinity, the amount concentrated depending on the fluoride content of the solution and the strength of the titrating solution. A better endpoint is obtained when less of the titrating solution is used. However, if the determination is to be made on a solid substance such as bone or rock phosphate, the material is finely ground, dried in the oven, and a sample weighed out, the weight of sample of course depending on the fluoride content. With finely ground and uniformly mixed samples of phosphate rock and bone containing



CROSS - SECTION

DISTILLING APPARATUS FOR DETERMINATION OF FLUORINE BY WILLARD & WINTER METHOD. FIG. 3

> 2% or more of fluorine, a 0.1 gram sample is sufficient. If the sample contains less than 2%, satisfactory results are usually obtained with a 0.2 gram sample.
>
> The sample is transferred to a distilling flask, 0.5 gram of pure powdered silica and 10 cc. of 60% perchloric acid is added. The flask is connected to the condenser and steam generator and heated to 80-90 degrees centigrade before admitting steam (see Fig. 3). The flow of steam is regulated so that 150 cc. of distillate is collected in 50-60 minutes; and the temperature in the flask is maintained at 135 degrees centigrade during the greater part of the distillation, the temperature being regulated by the inflow of steam and the electric heating unit by which the flask is heated. If a double-distillation is necessary, the first distillation is made with concentrated sulfuric acid, the same precautions being observed. This distillate is then concentrated to 50 cc. and the second distillation made with perchloric acid.
>
> The distillate is made alkaline with 2% sodium hydroxide, using either litmus paper or one drop of phenolphthalein as indicator. Evaporate to a suitable volume or take an aliquot and proceed with the titration as under the procedure with no interfering substances present.

Sanchis (49) also used alizarin red and zirconium nitrate as an indicator in his colorimetric method of determining fluorine in water. Elvove in 1933 (22) suggested that the zirconium-alizarin reagent of De Boer (17) might be utilized in this connection, and suggested a procedure with several modifications. Willard and Winter (67), however, stated that they attempted to use the colorimetric method based upon a zirconium-alizarin mixture, but that their results did not show any definite relation between the fluorine content and the fading of the color. On the other hand, Casares and Casares (11) utilized this reagent for determining fluorides in mineral waters, and Thompson and Taylor (65) applied it to the determination of fluorides in sea water.

The Sanchis method (49) is a modification of the Thompson and Taylor method. The direct application of this procedure to fresh waters did not seem practical because the wide fluctuation of their chloride to sulfate ratio, as well as the actual amount of these radicals present, almost necessitates the preparation of a set of standards for each sample to be analyzed. However, in the Sanchis modification the standards are made up with distilled water and chloride and sulfate effects minimized somewhat by the addition of hydrochloric and sulfuric acids.

This method was used whenever possible in the problem since it is relatively simple and quite reliable if the sulfate and phosphate concentrations are not too high.

The procedure as used in this work was very similar to that as proposed by Sanchis except that the two acids (3 N hydrochloric and 3 N sulfuric acids) were mixed and added together, and 250 cc. beakers were used instead of 250 cc. Erlenmeyer flasks as suggested by Sanchis. The procedure as used follows:

Sanchis Method

Reagents:

(1) Standard Sodium Fluoride Solution:

Prepare a stock solution containing 2.21 grams of C. P. dry sodium fluoride per liter. Dilute 10 cc. of stock solution to 1 liter (1 cc. equals 0.01 mg. of fluorine or 10 parts per million).

(2) Indicator:

Prepare a stock solution as follows: (a) dissolve 0.17 gram of alizarin sodium sulfonate (alizarin red) in 100 cc. of distilled water; (b) dissolve 0.87 gram of crystalline zirconium nitrate, C. P., in 100 cc. of distilled water. Add (a) to (b) slowly with constant stirring. Shake at intervals and allow to stand overnight. Dilute 20 cc. of stock solution to 100 cc. to serve as indicator. When not in use, these solutions are best kept in a cool dark closet.

(3) Hydrochloric acid, 3 N.

(4) Sulphuric acid, 3 N.

Preparation of Standards:

To each of a series of nine 250 cc. beakers, add 0, 2.5, 5.0, 7.5, 10.0, 15.0, 20.0, 25.0, and 30.0 cc. of standard fluoride solution (10 parts per million) and make up to a volume of 100 cc. with distilled water. These standards are treated in a manner identical to that prescribed for the unknowns below.

Procedure:

A 100 cc. aliquot of the sample to be analyzed, freed from turbidity and suspended solids by filtration if necessary, is placed in a 250 cc. beaker. Smaller aliquots may be taken depending on the fluoride concentration of the water, the sample being made up to 100 cc. with distilled water. To each 100 cc. aliquot thus measured, and to the prepared standards, add exactly 4.0 cc. of the mixed acid solution (3 N HCl and 3 N NaOH) and 2.0 cc. of indicator solution. The intensity and shade of the color produced depends upon the concentration of indicator and acids in each of the aliquots. It is, therefore, necessary to add identical amounts of these reagents to both standards and unknowns in order to obtain the maximum possible accuracy in the determination.

Place the beakers on an already heated hot plate. Bring the contents rapidly to the boiling point and remove as soon as boiling begins. Do not allow to boil vigorously nor to simmer for any length of time.

Four hours after cooling, or the following day, transfer the standards to properly labeled 100 cc. matched Nessler tubes and make up to the 100 cc. mark with distilled water. Transfer each of the unknowns, in turn, to a 100 cc. Nessler tube, make up to the mark, and compare its color with that of the standards.

If a reddish precipitate appears in any of the aliquots after cooling, disperse it by rapid whirling of the contents of the flask prior to their transfer to the Nessler tube and proceed with the determination as usual.

Investigation of Sanchis Method:

In Sanchis' original article (49) on the determination of fluorides in natural waters, he states: "The results of several hundred determinations, made with synthetic waters by the method here proposed, indicate that chlorides, sulfates, bicarbonates, sodium, calcium, and magnesium up to 500 parts per million; manganese up to 200 parts per million; silicates up to 50 parts per million, phosphates, boron, copper, and iron up to 5 parts per million; and sulfides up to 2 parts per million, do not interfere with the quantitative determination of fluorine".

However, it was noted that under certain conditions a coagulation of the dye (indicator) resulted on heating the sample, which rendered comparison of the sample with the standards in the Nessler tubes quite difficult. A study, therefore, was made on the effect of various ions on the reproducibility of results by this method. The study included phosphate ion, sulfate ion, chloride ion, calcium ion, and bicarbonate ion. Standard fluorine solutions were prepared and equilibrated with solutions of the above ions of different concentrations. Samples were then removed and the fluoride content determined by the Sanchis method. A comparison of the results thus obtained was then made with the values found for those solutions containing only fluorine.

The effect of the phosphate ion is shown in Table 1 and Fig. 4. It will be noted that as the phosphate concentration increases, the results obtained show a decrease; and the effect of the phosphate ion is more pronounced when the fluorine concentration becomes greater. For example, when the initial fluorine content was 0.5 per million, the addition of phosphate in an amount sufficient to bring its final concentration to 25 parts per million, showed a decreased reading of 0.4 of a part per million of fluorine; but when the initial fluorine content

THE USE OF BONE AND OTHER PHOSPHATES FOR THE REMOVAL OF FLUORINE FROM DRINKING WATER

INTRODUCTION

It is a well established fact that the presence of fluorides in water used for drinking and cooking purposes during the period of calcification of the teeth causes what is known as "mottled enamel", or more specifically, "chronic endemic dental fluorosis". Hitherto, water-borne diseases of endemic proportions have been considered to be due to pathogenic bacteria or other organisms; but through a series of intensive investigations starting about 1930, it has been established that the element fluorine combined as the fluoride, when present in the water supply in excess of 0.9 of a part per million, affects the teeth of growing children. This defect is most readily noted among the children of the grade school age in areas in which the fluorine concentration in the drinking water is above the "toxic" level of 0.9 of a part per million. A new and highly important problem of serious social and economic aspect immediately arises. How are these toxic quantities of fluorine to be removed from affected waters? What steps may be taken by the individual to prevent the occurrence of mottled enamel in his children?

An attempt is made in this paper to answer these questions through a study of the effectiveness of bone and other phosphates for use in removing fluorine from water.

was 2.0 parts per million, a decrease in reading of 0.9 of a part per million was obtained. However, when the concentration of the phosphate ion is under 10 parts per million, little change in the actual fluoride content is noted. This was chosen as the upper limit of phosphate concentration for use of the Sanchis method. Whenever coagulation of the indicator occurred, the water was analyzed for its phosphate content colorimetrically by means of ammonium molybdate and stannous tin. If the phosphate concentration exceeded 10 parts per million, the sample was reanalyzed using the Willard and Winter distillation procedure. This procedure was found necessary on only a few samples since the maximum solubility of the treated bone with respect to the phosphate ion is in the neighborhood of only 3 parts per million. However, on certain laboratory and field tests involving the synthetically prepared tricalcium phosphate, "defluorite," it was necessary to use the distillation method since the solubility of this material with respect to phosphate ion is much greater, often as high as 200 parts per million.

No explanation as to this reduction of the fluoride reading caused by phosphate ion can be readily advanced; but it may be due to a complex reaction involving the indicator, the phosphate, and the fluoride such that the fluorine is "tied" up as an insoluble form. The fluorine, thus being present as an insoluble complex, does not affect the indicator and results obtained are low. The actual coagulation seen is evidence that a side-reaction of some sort is taking place.

Scott in his paper on fluorides in Ohio water supplies (50) incorporates the following table of corrections for the presence of the sulfate

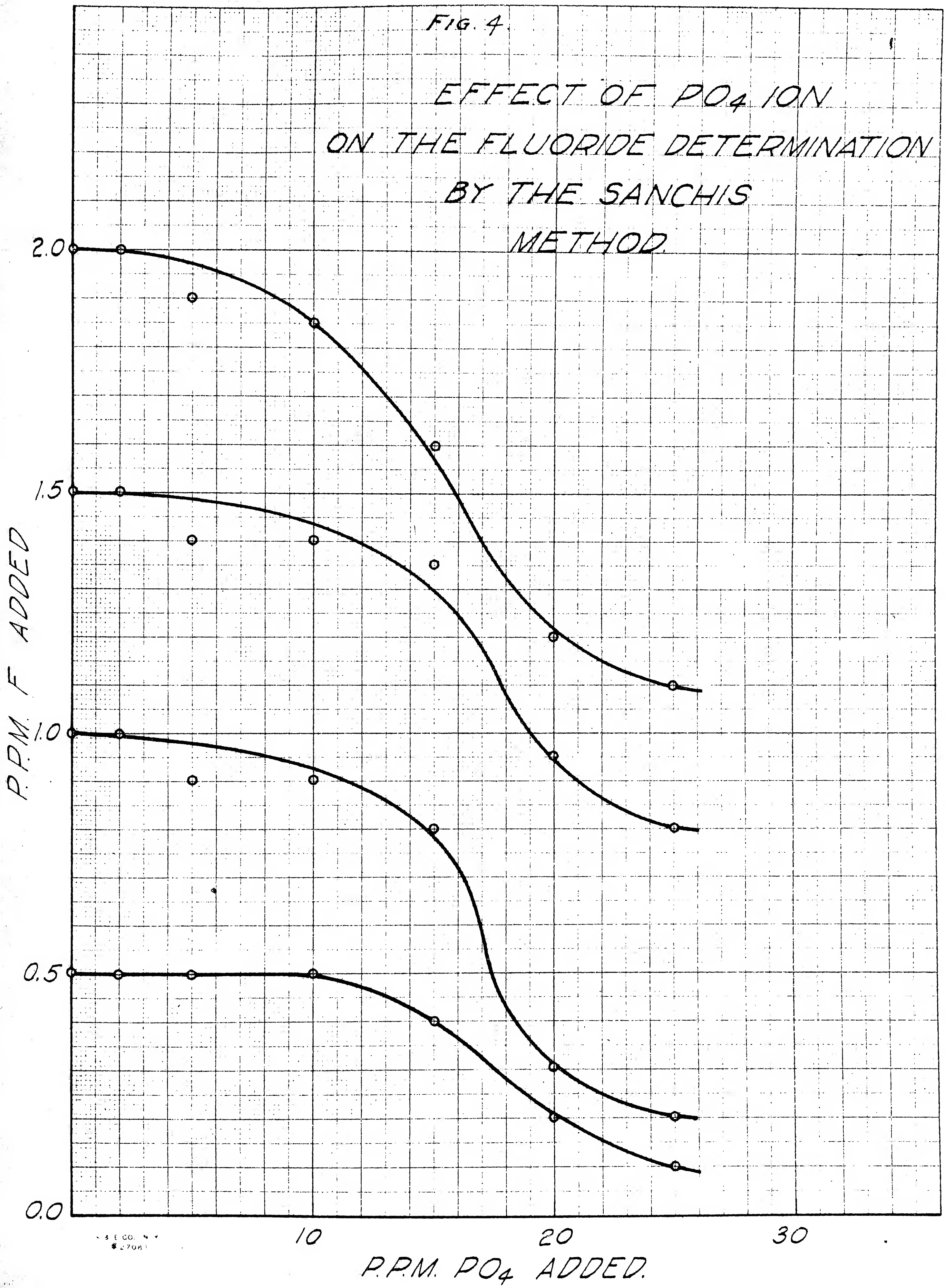
TABLE 1. EFFECT OF THE PHOSPHATE ION ON THE SANCHIS METHOD

Initial Fluorine	Phosphate Added	Final Fluorine
0.0 PPM	0.0 PPM	0.0 PPM
0.0	2.0	0.0
0.0	5.0	0.0
0.0	10.0	0.0
0.0	15.0	0.0
0.0	20.0	0.0
0.0	25.0	0.0
0.5	0.0	0.5
0.5	2.0	0.5
0.5	5.0	0.5
0.5	10.0	0.5
0.5	15.0	0.4
0.5	20.0	0.2
0.5	25.0	0.1
1.0	0.0	1.0
1.0	2.0	1.0
1.0	5.0	0.9
1.0	10.0	0.9
1.0	15.0	0.8
1.0	20.0	0.3
1.0	25.0	0.2
1.5	0.0	1.5
1.5	2.0	1.5
1.5	5.0	1.4
1.5	10.0	1.4
1.5	15.0	1.35
1.5	20.0	0.95
1.5	25.0	0.8
2.0	0.0	2.0
2.0	2.0	2.0
2.0	5.0	1.9
2.0	10.0	1.85
2.0	15.0	1.6
2.0	20.0	1.2
2.0	25.0	1.1

PO_4 added as $Na_3(PO_4)_2$
F added as NaF

FIG. 4

EFFECT OF PO_4 ION ON THE FLUORIDE DETERMINATION BY THE SANCHIS METHOD.



ion. If the sulfate, SO_4, concentration is as much as 250 parts per million in the tube as read, corrections are applied:

250	PPM	SO_4	0.02	PPM	apparent	fluoride,	F
300	"	"	0.05	"	"	"	"
400	"	"	0.10	"	"	"	"
500	"	"	0.15	"	"	"	"
700	"	"	0.20	"	"	"	"

The effect of sulfate ion was also included in this study and results obtained are shown in Table 2. It is seen that the differences in readings observed are in close agreement with the table as used by Scott and cO-workers. Why sulfate should give high results, is not known. It is possible, however, that sulfate ion may in some measure take the place of fluorine in the reaction.

The results of the study involving the effect of chloride, calcium, and bicarbonate ions on the method are shown in Table 2. It will be noted that these ions appear to have no effect on the accuracy of the method as indicated by the original work of Sanchis.

TABLE 2. EFFECT OF VARIOUS IONS ON THE DETERMINATION OF FLUORINE BY THE SANCHIS METHOD

Initial concentration of fluorine = 0.5 PPM
Fluorine present as NaF

Ion Added	Concentration of Ion Added (PPM)	Final Concentration of Fluorine (PPM)
Sulfate as Na_2SO_4	0	0.50
	100	0.50
	200	0.50
	500	0.70
	1000	0.90
Chloride as NaCl	0	0.50
	100	0.50
	200	0.50
	500	0.50
Calcium as $CaCl_2$	0	0.50
	25	0.45
	50	0.40
	100	0.50
	200	0.45
Bicarbonate as $NaHCO_3$	0	0.50
	50	0.50
	100	0.45
	200	0.45
	500	0.45

Preliminary treatment, activation, and regeneration of bone:

The bone used throughout the studies conducted was ordinary beef bone, usually obtained from the larger packing companies. Several of these establishments have been very cooperative in this respect, having furnished a large amount of bone of different sources and varieties (see acknowledgment). Without this help the extensive field studies which were carried on would have been somewhat limited.

In practically every case the bone received had been boiled or steamed with water to remove most of the fat, protein, and other organic materials, and had been ground to about quarter-inch mesh. In the laboratory the bone was further crushed and boiled with alkali (about 2 N) to remove more completely the organic material present. This treatment was continued until the bone had lost its flinty characteristics and had become snow white in appearance. The bone was then washed in water, dried, and graded in fineness. The graded fractions were again treated with alkali, washed with water, the excess alkali neutralized with hydrochloric acid (about 0.25 N), washed again with water, and finally dried. Bone treated in this manner now shows the capacity for removing fluorine from water.

The initial activation of the bone with alkali and acid suggested the possibility of regeneration of the material after its capacity for removing fluorine from water had been exhasted. Experiments conducted on this "spent" bone showed that the product could be made active once more by again treating with sodium hydroxide and hydrochloric acid just as in the initial preparation. The principle of the activation appears to be the same in each case -- a preliminary treatment with a base followed by a neutralization with an acid. It has been found that practically any base-acid combination will produce a rejuvenated bone. Experiments conducted with sodium hydroxide followed by a treatment with lactic, tartaric, oxalic, citric, phosphoric, boric, formic, acetic, propionic, carbonic, hydrochloric, sulfuric, and nitric acids all have produced a bone of practically the same fluoride removing power. Results of this study are shown in Table 3.

Potency value:

Early in the course of this problem it was realized that a certain definite standard treatment of the bone products obtained from the various experiments conducted, was necessary to compare one product with another. For this reason a test was chosen which has become known in the laboratory as the "standard potency test". This test consists in shaking 2 grams of the material for one hour in and end-over-end shaker (15 R. P. M.), with 500 cc. of a 3 part per million solution of fluorine made up in distilled water. The mixture is immediately filtered and the residual fluorine determined on the filtrate, usually by the Sanchis method. By using this purely arbitrary method a comparison of products may be made in the laboratory. The so-called "potency" value, as it is defined in this paper, refers to the residual fluorine left in the solution after the shaking has taken place.

TABLE 3. EFFECT OF REGENERATION OF BONE WITH VARIOUS ACIDS ON POTENCY VALUE (40 - 60 MEST BONE)

Potency values in p.p.m. fluorine

	Time of Contact (hours)		
Acid Used	$\frac{1}{8}$	2	24
Lactic	0.40	0.40	0.40
Tartaric	0.45	0.35	0.40
Oxalic	1.10	0.90	1.00
Citric	0.40	0.50	0.45
Phosphoric	0.50	0.45	0.50
Boric	0.60	0.65	0.60
Formic	0.40	0.45	0.40
Acetic	0.40	0.35	0.45
Propionic	0.40	0.40	0.50
Hydrochloric	0.50	0.60	0.50

Referring to Table 3 again, it is found that the sodium hydroxide treatment followed by neutralization with the various acids mentioned above results in a bone product of practically the same fluoride removing value or "potency"value in each case. However, regeneration in nearly all of the above mentioned cases was accomplished only with a 15 to 25 per cent loss of bone. This dissolution loss led to a study of the minimum concentrations of alkali and acid necessary for reactivation as well as the conditions best suited to a rejuvenation of the bone in an effort to "cut" down the percentage loss.

A series of experiments was first set up to study the minimum concentration of acid that could be used. A quantity of bone was boiled for several hours with 0.5 N sodium hydroxide solution, washed, and dried. Samples of this material were treated with acetic acid ranging in concentration from 0.001 N to 0.2 N, and with hydrochloric acid ranging in concentration from 0.05 N to 0.2 N. The time of contact of acid and bone was varied and the percentage loss noted in each case. Results of this study are shown in Table 4. The results show that the strength of acid used is not important so long as sufficient acid is present to neutralize the excess alkali used in the preliminary treatment. The potency values obtained with the more dilute concentrations are practically the same as with the higher concentrations. Any excess acid seems to react with the carbonate of the bone thereby creating a greater percentage loss on regeneration. The time of contact of the bone and the acid, also, does not appear to be of importance so long as sufficient time is allowed for neutralization to occur. A 5 to 10 minute treatment with acid has been found to produce a reactivated bone and at the same time reduce the loss due to dissolution.

The second step in this study was the determination of the strength of alkali necessary for rejuvenation of the bone. It has been found that the sodium hydroxide treatment is the most important step in the reactivation process. A series of experiments was set up using bone which had lost its capacity for removing fluorine. Samples of this material were treated with various concentrations of alkali for different periods of time. The experiment was conducted by boiling the bone with the alkali in one case and by treatment in the cold in the second case. The percentage loss on alkali treatment, the total percentage loss on alkali and acid treatment, and the potency value of each product was also determined. Neutralization of the excess alkali was accomplished in each case by treatment with 0.1 N acetic acid for 10 minutes. The results of the experiment are shown in Tables 5 and 7.

It appears that reactivation with the sodium hydroxide in the cold results in a product just as good as that obtained by boiling with the alkali, and, in addition, the solubility loss is less. In marked contrast with the acid treatment, however, it was found that the strength of base used and the time of contact necessary was much greater. But, since the solubility loss in the cold with the alkali is much less than that obtained with the acid, a twenty-four hour treatment with a stronger base is not out of the question. The added fluoride-removing power of bone treated for longer periods of time with greater strengths of base more than make up for the solubility losses which may occur. The total percentage loss for reactivation in the cold with 0.2 N NaOH and 0.1 N acetic acid has been found to be from 10 to 12 per cent. This loss by dissolution may be

TABLE 4. EFFECT OF STRENGTH OF ACID ON REGENERATION OF BONE.
(40 - 60 MESH BONE)

Acid Used	Concentration of Acid Used (Normality)	Time of contact of Bone and Acid (Mins.)	Percentage loss (%)	Residual F Potency (p.p.m.)
Acetic	0.001	5	3.5	1.1
		10	7	1.0
		30	9	0.9
	0.005	5	7	0.9
		10	10	0.9
		30	11	0.9
	0.01	5	8	0.9
		10	9	0.9
		30	10	0.9
	0.05	5	8	0.9
		10	10	0.9
		30	14	0.9
	0.10	5	10	0.9
		10	13	0.9
		30	14	0.9
	0.20	5	17	0.9
		10	20	0.8
		30	22	0.8
Hydrochloric	0.05	5	17	0.9
		10	18	0.9
		30	20	0.9
	0.10	5	15	0.9
		10	18	0.8
		30	21	0.8
	0.20	5	17	0.8
		10	21	0.8
		30	23	0.8

TABLE 5. EFFECT OF VARYING ALKALI CONCENTRATION ON THE REGENERATION OF BONE BY BOILING WITH SODIUM HYDROXIDE.

(40 - 60 MESH BONE)

Strength of NaOH Used (Normality)	Time of Treatment (Minutes)	Percentage loss from NaOH (%)	Total Percentage loss alkali and acid* (%)	Residual F Potency (p.p.m.)
0.01	5	3.6	14.0	2.7
	15	6.4	10.8	2.5
	30	4.0	11.2	2.0
	60	7.0	11.6	2.2
0.05	5	3.6	9.2	2.0
	15	2.4	15.6	2.0
	30	2.8	14.0	1.7
	60	5.2	18.8	1.7
0.10	5	5.6	12.8	1.6
	15	7.0	12.5	1.7
	30	8.0	14.0	1.6
	60	10.0	15.6	1.5
0.20	5	7.2	12.8	1.4
	15	7.6	13.2	1.3
	30	8.0	11.2	1.4
	60	10.4	18.8	1.3

*Neutralization of the excess base was made by treatment with 0.10 N acetic acid for ten minutes.

TABLE 6. EFFECT OF CARBON-DIOXIDE GAS ON REGENERATION OF BONE.

(40 - 60 MESH BONE)

Treatment	Total Percentage loss	Residual F p.p.m.
Boiled with NaOH--washed with water--carbon-dioxide passed 15 minutes	4.9	0.9
Treated with NaOH in cold for 20 hours--washed with water--carbon-dioxide passed 15 minutes	1.8	1.0

TABLE 7. EFFECT OF VARYING ALKALI CONCENTRATION ON THE REGENERATION OF BONE IN THE COLD WITH SODIUM HYDROXIDE.

(40 - 60 MESH BONE)

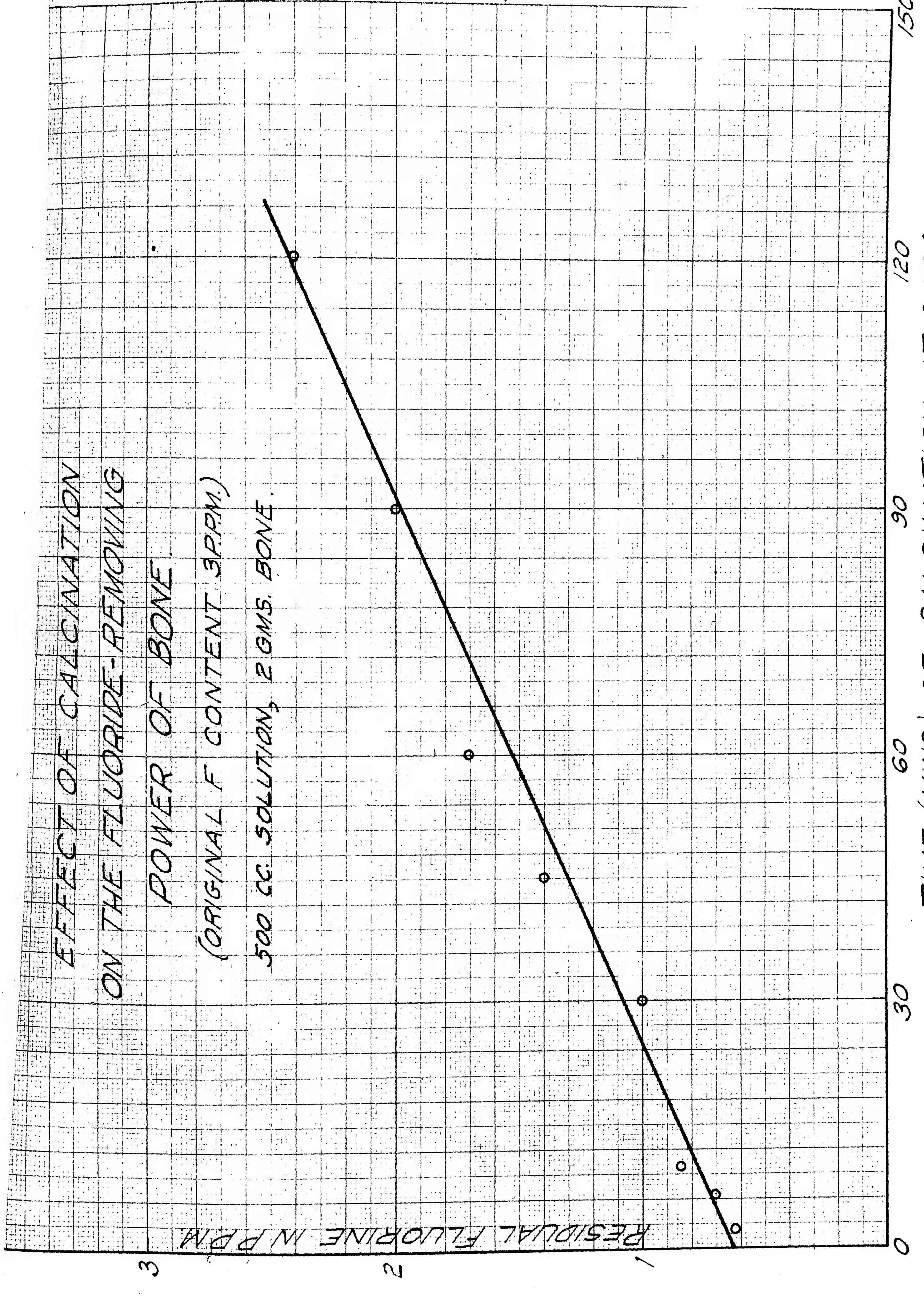
Strength of NaOH Used (Normality)	Time of Treatment (Minutes)	Percentage Loss from NaOH (%)	Total Percentage loss alkali and acid* (%)	Residual F Potency (p.p.m.)
0.01	5	0.0	8.0	2.8
	15	1.2	7.6	2.5
	30	2.3	9.2	2.5
	60	6.0	11.6	2.4
0.05	5	1.2	10.8	2.0
	15	2.0	10.4	1.9
	30	4.8	14.4	1.8
	60	3.2	14.0	1.8
0.10	5	0.4	6.2	2.4
	15	5.2	13.2	2.5
	30	8.0	14.4	2.0
	60	4.8	14.0	1.6
0.20	5	2.8	7.6	1.8
	15	6.4	11.2	2.0
	30	6.0	13.2	2.0
	60	6.8	12.9	1.5
0.01	16 hrs.	1.0	4.6	2.2
0.05	16	2.0	7.2	1.6
0.10	16	2.4	10.0	1.3
0.20	16	2.8	12.0	0.9

*Neutralization of the excess base was made by treatment with 0.10 N acetic acid for ten minutes.

somewhat decreased, however, by using carbon-dioxide gas for the purpose of neutralization. Experiments conducted using this method have shown that the average loss may be decreased to from 1 to 2 percent.

Results showing the neutralization of the sodium hydroxide-treated bone by bubbling carbon-dioxide gas through a suspension of the material are shown in Table 6, together with the potency value obtained and the percentage loss on reactivation. Since the product obtained under this treatment is as effective in removing fluorine as that obtained by using stronger acids and the percentage loss is markedly decreased, this procedure is recommended for reactivation purposes.

FIG. 5.



EFFECT OF CALCINATION ON THE FLUORIDE-REMOVING POWER OF BONE

(ORIGINAL F CONTENT 3P.P.M.)

500 CC. SOLUTION, 2 GMS. BONE.

Calcination studies:

It is recognized that the removal of fluorine by bone is a surface reaction, and that if the surface is increased, the effectiveness of the bone in removing fluorine from water is likewise increased. A greater surface area may be obtained by grinding the material, but this procedure has a serious limitation; namely, the flow of water through the finely ground bone is decreased so much that the use of this material becomes impractical in most types of filters except perhaps certain pressure filters. Another procedure commonly used for increasing surface area without decreasing the state of subdivision of the bone is the process known as calcination. This merely involves the heating of the bone at a high temperature for a short period of time. Calcination removes the volatile material in the pores of the bone and causes a certain amount of decrepitation which has the effect of increasing the surface area of contact.

A number of samples of alkali-acid activated bone (20-40 mesh) were heated in silica crucibles at different temperatures and standard potency tests made on the products. The results are shown in Table 8. An examination of the data shows that calcination of bone for short periods of time effects little change in the fluoride-removing power, but that on prolonged heating, especially at higher temperatures, a marked decrease in the "potency" of the bone is obtained (see Fig. 5). It was thought that perhaps a more uniform heating together with the constant supply of moisture would be desirable and a better product would be obtained. For this purpose a small rotary kiln was constructed. This apparatus was so

TABLE 8. EFFECT OF CALCINATION ON THE FLUORIDE REMOVING POWER OF BONE.

Treatment	Temperature of Calcination C	Time of Heating (Minutes)	Residual F Potency (p.p.m.)
Original bone	---	---	0.60
Calcination in air	200	2½	0.85
		5	0.75
		10	0.70
		30	0.65
		45	0.65
		60	0.65
		120	0.60
	400	2½	0.60
		5	0.60
		10	0.60
		30	0.70
		45	0.70
		60	0.70
		120	1.10
	600	2½	0.62
		5	0.70
		10	0.85
		30	1.00
		45	1.40
		60	1.70
		90	2.00
Calcination with steam	350	10	1.40
	400	10	1.50
	600	2	1.20
		5	1.25
		10	1.35
		15	1.60

designed that it would fit into an electric muffle and could be rotated constantly throughout the heating process. Steam was supplied continuously to the system through a hole in the muffle door. The kiln was placed in the furnace, brought to the desired temperature, and the bone then added and heated with constant rotation for the predetermined period of time. Standard potency tests conducted on the material prepared in this manner were disappointing (see Table 8). The bone products did not show as great a fluoride removing power as bone treated in the dry state and, hence, this steam process was abandoned as an unnecessary step in the preparation of the bone.

It was thought that possibly during the heating process the bone was converted to the oxy- or hydroxy-apatite and that, therefore, a treatment with hydrochloric acid was necessary to render the bone potent again. Consequently, several representative portions of the calcined bone were treated with dilute hydrochloric acid and standard potency tests made on the products obtained. Results of the treatment are shown in Table 9.

TABLE 9. EFFECT OF ACID TREATMENT ON CALCINED BONE

(40-60 MESH BONE)

Calcination Treatment	Acid Treatment	Residual F p.p.m.
10 min. at 200° C.	20 hours with 0.2 N HCl	0.50
2 hours "	"	0.70
10 min. at 400° C.	"	0.40
2 hours "	"	0.60
10 min. at 600° C.	"	0.40
2 hours "	"	0.70
Unheated bone		0.60

Historical review:

The first record of mottled enamel was apparently made at Naples, Italy in 1901 by Surgeon J. M. Eager (20) of the U. S. Public Health Service who noted this condition in the teeth of emigrants.

In the United States, F. S. McKay and C. V. Black (40) made the first recorded observations of mottled enamel in 1916. The first definite recognition of the relation of water to mottled enamel was given by McKay (41) in 1926.

Oakley, Idaho in 1925 (19) and Bauxite, Arkansas in 1930 (67) were forced to develop new water supplies because of endemic mottled enamel.

The fact that fluorides in drinking water are the cause of this dental defect was first definitely correlated by M. C. Smith, E. M. Lantz, and H. V. Smith (64) at the University of Arizona in 1931. These investigators were able to prove that waters containing fluorine in excess of 1 part per million were definitely associated with the occurrence of mottled enamel; and, furthermore, they produced mottled enamel on the teeth of rats by incorporating various amounts of sodium fluoride in the diet. The further researches of Smith and Smith (56,57,58,59),Smith, H. V. (54,55), Smith, M. C. (60), Smith and Lantz (61,62), Leverton and Smith (36), Schour and Smith (51), and the recent work of Churchill, (12), McKay (42), Kehr (31), Ostrem, Nelson, Greenwood and Wilhelm (43), Boissevain (7), Dean (16), Sebrell, Dean, Elvove and Breaux (52), and Boruff and Abbot (8) have contributed greatly to our knowledge of the distribution of fluorine in drinking waters and the effect of such waters in the production of mottled enamel of the teeth.

The acid after-treatment produces a slightly more potent bone, when applied to bone heated for 10 minutes; calcination for 2 hours followed by the acid after-treatment was, however, without effect. Over the range of temperatures studied it is to be noted that the best calcined products are those that have been heated at the higher temperatures for short periods of time.

Further study has shown that short time treatments with acid are as effective as the twenty-hour treatment used in the first study (Table 13). Results of these "flash" acid treatments are shown in Table 10.

TABLE 10. EFFECT OF SHORT-TIME ACID TREATMENTS ON THE FLUORIDE REMOVING POWER OF CALCINE BONE.
(40 - 60 MESH BONE)

Time of Calcination at 600° C. (Minutes)	Acid Treatment	Residual F Potency (p.p.m.)
10	5 min., 0.1 N HCl	0.50
	15	0.60
	30	0.60
	60	0.50
	180	0.65
	5 min., 0.2 N HCl	0.60
	15	0.75
	30	0.80
	60	0.80
	180	0.75

Calcination of bone for 10 minutes at 400 to 600 degrees centigrade followed by a 5 to 15 minute acid treatment has been found to yield a good bone product for the fluoride removal process.

It was found by Reynolds, Jacob, and Rader (44) that in the presence of sufficient silica and water vapor, heating phosphate rock for 30-60 minutes at 1400 degrees centigrade results in the volatilization of

95-100% of the fluorine and conversion of 85-95% of the phosphorus into the citrate soluble condition. Since nearly all of the fluorine is driven off in the process, it was thought that perhaps the rock would function in removing fluorine from water. Hence, a sample of the material was obtained from Mr. Jacob and several trials made. Potency tests conducted on the defluorinated rock phosphate were disappointing (see Table 11). Treatment of this material with alkali and acid as used on the activation of bone also gave unsatisfactory results.

TABLE 11. EFFICIENCY OF DEFLUORINATED ROCK PHOSPHATE IN REMOVING FLUORINE FROM WATER. (20 - 40 MESH)

Weight of Rock (Grams)	Mesh	Initial F (p.p.m.)	Residual F (p.p.m.)
2	20-40	3.0	2.3
10	20-40	3.0	2.2

The greater fluoride-removing power which calcination gives the bone would hardly warrant the cost of this additional step, however, if this procedure had not been found necessary for the preparation of a suitable material for use in a filter. There are two reasons why this process has been found necessary -- (1) it makes bone more easily "wettable"; and (2) it prevents microbial growth in the filter.

It was noted that on certain occasions the bone activated by the usual alkali-acid treatment refused to measure up to expectations in regard to the fluoride removing power. This decrease in potency was found to be caused by the non-wetting of the bone. Often the prepared material would float on the top of the water. In other instances the bone would seemingly be "wetted" but on closer observation it was noticed that each bone particle was enclosed in a tiny air bubble. Obviously under these conditions the effectiveness of the bone was reduced. The reason for this "non-wettability" of the bone may probably be explained on the basis of the presence of an oil film around the bone particles. The treatment which is given the bone would not be expected to remove all organic compounds present. It is most likely that these bone products which refuse to "wet", have a flim, perhaps only monomolecular, of oil surrounding each particle, and, hence, the reason for the non-wetting. The calcination process evidently destroys this film, since all products prepared in this manner have "wetted easily and quickly.

Studies on the putrefaction of bone:

The second reason for the necessity of calcination, that of preventing microbial growth to develop in the bone, is more important, and would alone more than make up for the added cost of this procedure. A number of laboratory and field studies have shown that uncalcined bone has a tendency to putrefy, and thus to impart a disagreeable odor and taste to the water. This effect generally appears after the filter has been used for some time and is then allowed to remain inactive for several days. Also moist samples of bone left undisturbed in the laboratory have produced evidences of putrefaction.

A few attempts made to isolate the responsible organism or organisms proved unsuccessful. The organisms, however, have been shown to be anaerobic or semi-anaerobic since the putrefaction develops only in bone covered with water or in the lower portion of a moist column of bone. The microorganisms and mal-odor which they develop may be quickly destroyed by exposure to light and air and allowing the bone to dry. Preliminary results indicate that some type of mold is responsible since only those organisms which grew on mold media produced a similar growth when inoculated into bone. Various types of culture media were prepared and inoculated with a suspension of the infected bone and with the infected bone itself. The typical organisms which developed were reinoculated into Noguchi tubes filled with moist sterile bone. The Noguchi tube is a long narrow test tube specially designed such that when filled with moist bone, anaerobic conditions develop in the lower portions, and aerobic conditions towards the top of the tube. The lower portions of the bone in the Noguchi tubes, when inoculated by means of a long needle with the material immediately surrounding the bone on the mold media, produced evidences of similar growth as was noted in the field units and in the laboratory. Putrefaction of sterile bone also could be brought about by covering the bone with water and inoculating with the infected material. Sterilization of the bone with a number of common sterilizing agents such as hydrogen peroxide, chlorox, and hychlorite were tried, and, although these agents would quickly clear up the condition on the first application, putrefaction would again develop after use of the bone. However, calcined bone has never shown any tendency to putrefy. Inoculations which have quickly produced putrefaction in the case of uncalcined bone, have never caused any growth of microorganisms to develop on calcined bone. The results of a few of such inoculations are shown in Table 12.

TABLE 12. EFFECT OF INOCULATIONS OF CALCINED AND UNCALCINED BONE

Source of Bone	Treatment	Time of Inoculation	Time of Putrefaction
Eagle Milling Co., Tucson, Arizona	Alkali-acid-calcined	2/1/39	No growth at 5/7/39
"	Alkali-acid-calcined	2/1/39	No growth at 5/7/39
"	Alkali-acid-no calcination	2/15/39	2/18/39

Extensive field tests have been carried on with calcined material and, as yet, not one case of putrefaction has occurred.

Why calcination prevents the growth of microorganisms on the bone may in all probability be explained on the basis of the change which the organic matter content undergoes at these temperatures. The organic matter at these high temperatures is probably changed by pyrolysis, dehydration, and oxidation to a form which will not support the growth of microorganisms.

In addition to rendering the bone wettable and non-putrefactive, there are several added advantages of calcination: (1) the bone is made harder and less subject to losses by attrition and dissolution; (2) the surface area is increased, thus increasing the capacity of removal; and (3) more fluorine is removed from the bone (see chemical analysis) which also increases its fluoride removing power.

Chemical analysis of bone:

It has long been known that the CaO: P_2O_5 ratio in bone is greater than that required for tricalcium phosphate (2, 26, 27, 34, 35, 45). There has long been a controversy as to the chemical composition of bone. Klement and Tromel (34) give the formula of bone as $3Ca_3(PO_4)_2 \cdot Ca(OH)_2$ plus $CaCO_3$. Gassmann (27) gives evidence that the structural part of bone is as Werner suggested -- $3Ca_3(PO_4)_2 \cdot CaCO_3$. Bassett (5) believed that the formula was probably $Ca_{10}(OH)_2(PO_4)_6$, with amorphous or crystalline $CaCo_3$. The The X-ray observations of DeJong and others (18,47) showed that the principal constituent of bone was an apatite-like substance. Hendricks, Hill, Jacob, and Jefferson (29), as a result of microscopical and X-ray diffraction examinations, report that bone is probably essentially a carbonate apatite and that for structural reasons it is probably $Ca_{10}CO_3(PO_4)_6 \cdot H_2O$. Several representative samples of bone were analyzed, not for the purpose of determining a formula, but to obtain an idea of the true bone content of the product. It was found that the values obtained for the percentage of CaO and P_2O_5 were lower in every case than the calculated theoretical percentages, assuming the formula to be that of carbonate-apatite or the hydrated carbonate-apatite as suggested by Hendricks. This may probably be explained by assuming that the treatment given the bone in the course of its preparation is not rigorous enough to remove all impurities present -- a certain amount of organic matter and probably some silica is present. The very fact that a loss of 40% of the bone material occurs during its initial preparation and activation serves as evidence that the steamed bone meal obtained from the packing houses is very unpure in nature. Even calcination does not remove all of the organic matter. However, a sufficient amount of the impurities has been removed to render the product safe for use in a bone filter. The results of the analysis for calcium, phosphorus, and fluorine are shown in Table 13, together with theoretical values calculated for several suggested formulas for bone and for fluorapatite.

TABLE 13. CHEMICAL ANALYSIS OF BONE. (DRY BASIS)

Type of Bone and Treatment Received	CaO %	P_2O_5 %	Ratio $CaO:P_2O_5$	F %
Raw chipped bone untreated	24.30	31.09	0.782	0.313
Chipped bone (alkali-acid)	42.84	40.28	1.063	0.228
Chipped bone (alkali-acid-F sat'd.)	41.58	36.44	1.144	1.912
Chipped bone (alkali-acid-calcined-acid treated)	41.72	37.09	1.124	0.174
Theoretical--$Ca_{10}(CO_3)(PO_4)_6$	54.33	41.27	1.316	
Theoretical--$Ca_{10}(CO_3)(PO_4)_6 \cdot H_2O$	53.39	40.56	1.313	
Theoretical--$Ca_{10}F_2(PO_4)_6$	54.56	42.06	1.297	3.76

The ratio of $CaO:P_2O_5$ should be the same approximately as that calculated from the theoretical percentage regardless of impurities present unless such impurities present in the bone are in the form of some other calcium and phosphorus compound. However, results show that the $CaO:P_2O_5$ ratio is lower in each case, indicating that more calcium is being dissolved during the activation and preparation of the bone.

A number of bone samples from different sources as well as several rock phosphates were analyzed for fluorine in order to determine the average fluorine content of these materials both in the natural state and in the treated form. It will be noted that the alkali-acid treatment results in a decrease of the fluoride content and that calcination results in a still further decrease. The rock phosphate samples show a high content approaching the theoretical of fluor-apatite. Also while saturation with fluorine under the conditions of the experiment (equilibrating with a high NaF concentration for several days) did not reach the value calculated from fluorapatite, the fluoride content was substantially higher than that of the treated bone. Results of the analysis are shown in Table 14. It will be noted that the fluoride content of bone after use in a filter has increased somewhat.

TABLE 14. FLUORINE CONTENT OF BONE AND ROCK PHOSPHATE. (DRY BASIS)

Material and Treatment	F %
Fla. rock phosphate (raw)	3.385
Fla. rock phosphate (calcined)	3.770
Fla. rock phosphate (raw-alkali-acid treated)	3.360
Fla. rock phosphate (Calcined-alkali-acid)	3.695
Steamed bonemeal poultry feed (untreated)	2.363
Apatite	0.075
Calcined bone (F saturated)	0.623
Calcined bone (calcined moist-acid treated)	0.205
Calcined bone (calcined moist-F saturated)	0.720
Calcined bone (after use in filter-capacity lost)	0.396

A number of different bone samples was analyzed for their protein content (nitrogen x 6.25) in order to determine in some measure the purity of the bone and its susceptibility to putrefy, it being assumed that the protein content would parallel the rate and degree of putrefaction. It was found that the alkali-acid treatment substantially reduced the organic matter content. However, calcination reduced the protein only slightly more than the alkali-acid process. As stated before the small amount of organic matter left in the bone is probably changed in form or the quantity becomes negligible, and, hence, no putrefaction occurs. Results of the analysis are shown in Table 15.

TABLE 15. PROTEIN CONTENT OF BONE. (AIR DRY BASIS)

Material	Source	Treatment	Protein N x 6.25 (%)
Steamed bone meal	Consolidated Chemical Industries, San Francisco, Cal.	alkali-acid	0.42
Chipped bone	Swift and Co., Ft.Worth Texas	alkali-acid	0.66
Raw bone	Swift and Co.,St.Paul, Minn.	alkali-acid	0.91
Calcined bone	Eagle Milling Co., Tucson, Arizona	alkali-acid-calcined-acid	0.47
"Digesta" bone	Glendale, Ariz.	alkali-acid	0.41
Steamed bone meal	Eagle Milling Co., Tucson, Arizona	alkali-acid	0.56
Steamed Bone "A"	Glendale, Ariz.	alkali-acid	0.84
Steamed bone "B"	Glendale, Ariz.	alkali-acid	0.81

Effect of mesh on fluorine removal and flow rates:

The effect of fineness of subdivision of the bone with respect to the amount of fluorine removed was investigated, and, as expected, was found to play an important role. A quantity of bone was prepared by the alkali-acid method and segregated into various meshes. Two-gram portions of the different sizes were shaken with 500 cc. of a 5 part-per-million fluorine solution for one hour in an end-over-end shaker (15 Revolutions Per Minute). The mixtures were immediately filtered and the filtrates analyzed for their residual fluoride content by the Sanchis method. Results of the experiment are shown in Table 16.

TABLE 16. EFFECT OF BONE MESH ON FLUORINE REMOVAL.

Bone Mesh	Weight of Bone Used (Grams)	Initial F Content (p.p.m.)	Final F Content (p.p.m.)
10-40	2	5.0	1.2
20-40	2	5.0	0.7
40-60	2	5.0	0.4
60-80	2	5.0	0.3
80-100	2	5.0	0.0
less than 100	2	5.0	0.0

The finer fractions of bone effected a much greater removal of fluorine, indicating that the reaction is a surface phenomenon. However, a finer mesh than 40-60 is considered impractical for use in the bone filter since the rate of flow becomes too slow. Also loss on regeneration of the bone becomes higher with decrease in the size of particles. A finer mesh than 40-60 may possibly be used in the pressure type of filter. Or a combination of meshes such as 40-80 may be used in the gravity type filter without hindering the penetration of water too much.

In connection with mesh studies the flow rates have been also investigated, the problem being approached from two directions-- (1) flow

Mottled enamel:

Mottled enamel, a term first used by Black, appears as a brown or yellow stain, at times absolutely black or in other cases as an abnormal opaque white. The condition is readily noted when the lips are open. It is chiefly characterized by the presence of dull chalky-white or paper-white patches distributed irregularly over the surface of the teeth. In many cases the whole tooth surface presents this dead-white, unglazed appearance. The teeth glare unnaturally when the mouth is open because of the loss of their normal translucency. Frequently the enamel is badly pitted and corroded, and the teeth are structurally weak, the enamel tending to chip off.

Mottled teeth may or may not become stained later. The stain is frequently confused with the enamel defect itself, but it is a secondary phenomenon. It shows considerable variation in coloration from dark brown, almost black, to yellow, and is usually not present on all of the teeth but more pronounced on the upper central incisors. The general tendency for the stain to follow the lip line suggests that exposure to air and light may be a factor in its production.

Histological examinations of mottled teeth by G. V. Black and others show that the intercementing material normally present between the enamel rods is lacking. In severe cases the enamel rods themselves are imperfectly calcified. The interference with the normal development of the enamel in all probability occurs during the period of its formation, for the first tip of a tooth seen breaking through the gum presents the typical white, unglazed appearance. Normally formed teeth have never been known rate as a function of mesh; and (2) flow rate as a function of depth of column of bone and the ability to remove fluorine from water.

The first experiments conducted involved the measurement of the rate of flow of water through a twelve-inch depth of bone of various screen sizes, the "head" of water being kept constant at twelve-inches in each case. For this study a long narrow tube of 2.25 cm. diameter was used. The results are shown in Table 17. As was expected the flow rate decreased markedly with the mesh of bone used.

TABLE 17. RATE OF FLOW OF WATER THROUGH BONE.

Bone Mesh	Flow Rate Gal./hr./sq.ft./12 in. column
10-20	4190
20-40	712
40-60	427

The second study conducted on this problem was of a more practical nature. The rate of flow and the fluorine absorbing power of bone was investigated as a function of the depth of column. Three glass tubes of varying diameters were chosen for the work, the first having a diameter of 0.55 inch, the second 0.86 inch, and the third 1.33 inches. In the first experiment the tubes were all filled with the same amount of bone (20 grams) and a solution containing 10 parts per million of fluorine passed through as rapidly as possible by gravity flow, a constant head of water of about twelve inches being maintained in each case. The rate of flow together with the amount of water treated was determined. This latter was determined by sampling the water passed through after every

TABLE 18. EFFECT OF FLOW RATE ON FLUORINE REMOVAL. (40 - 60 MESH BONE)

Conditions Varied	Diameter of Tube (inches)	Height of Column (inches)	Weight of Bone (grams)	Flow Rate Gal./sq.ft./hr.	Total Volume of 10 p.p.m. F treated Gal./sq.ft.	Grams F Removed per cu. ft. of bone
Height of column and diameter of tube	0.55	6.42	20	320	345.5	55.5
	0.86	2.57	20	300	199.2	50.1
	1.26	1.26	20	703	21.6	8.3
Weight of bone used	0.55	9	30	256	761.8	81.6
	0.86	9	72	248	1160.1	81.0
	1.33	9	169	219	1466.1	80.2

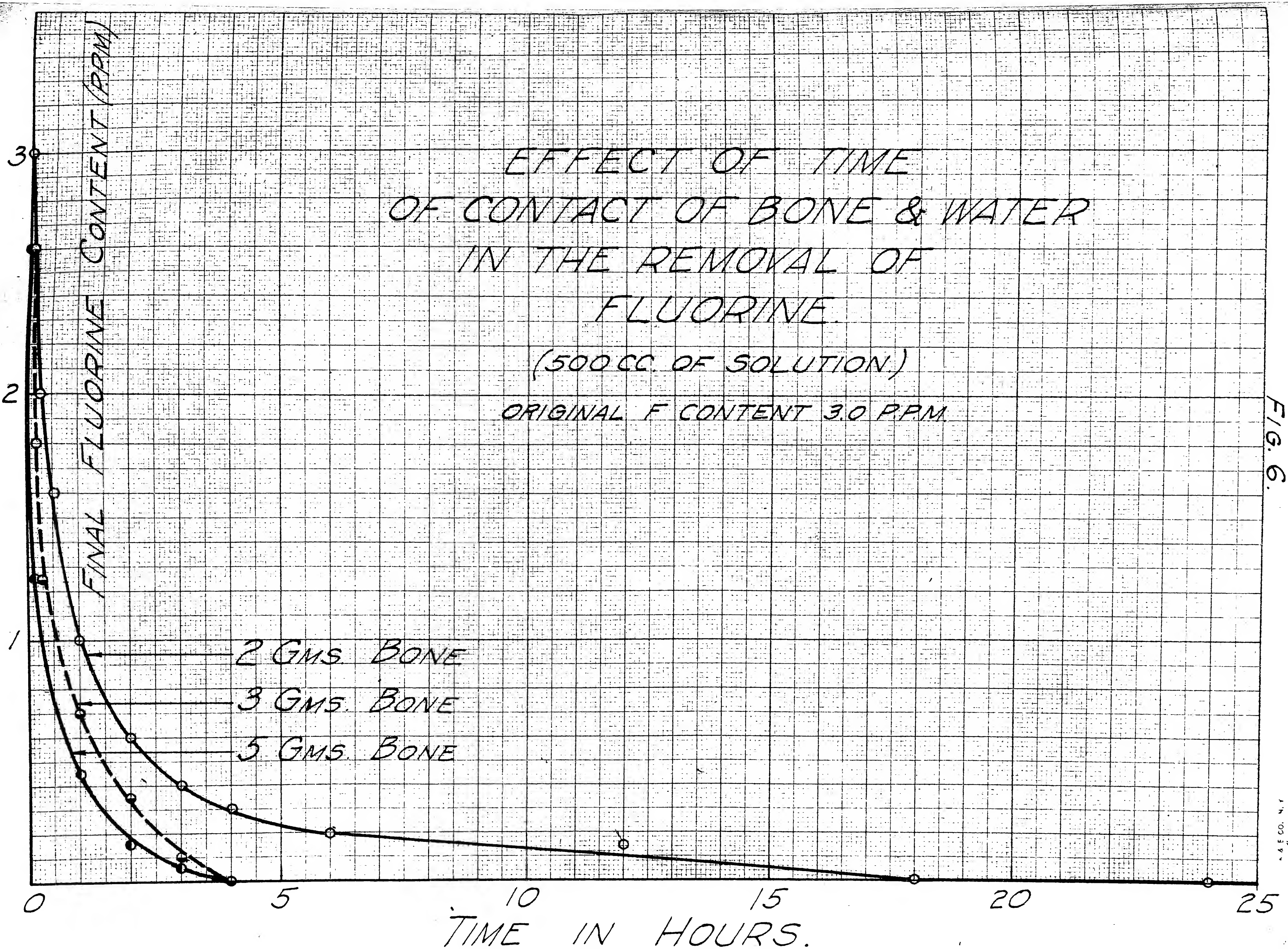
500 cc. and the residual fluorine content found by using the Sanchis method. Water was passed until the "toxic" limit of 0.9 part per million of fluorine was reached.

The results of the experiment are shown in Table 18. As was expected, the tube of smallest diameter and greater length of column had the slowest flow rate and, therefore, removed more fluorine from the water. The rate of flow through the large tube was so fast that only a very little fluorine was removed from the solution. The second experiment consisted of filling all three tubes to the same height with bone (nine inches) and noting the amount of water treated and the rate of flow in each case. Results of this study are also shown in Table 18. It will be noted that when the height of bone is the same in all three tubes, the total volume of water treated is not a function of the rate of flow as in the case of Table 21, but of the total weight of bone present. Also it is of interest to note that the amount of fluorine removed in each case is practically the same when calculated in terms of removal per cubic foot of bone.

TABLE 19. EFFECT OF TIME OF CONTACT ON THE REMOVAL OF FLUORINE FROM WATER BY BONE. (20 - 40 MESH BONE)

Time of Shaking	Weight of Bone used (Grams)	Initial Fluorine (p.p.m.)	Final Fluorine (p.p.m.)	Phosphate Content (p.p.m.)	Final pH value
1 min.	2	3.0	3.0	----	----
5	2	3.0	2.6	2.04	7.17
15	2	3.0	2.0	2.24	7.10
30	2	3.0	1.6	2.88	7.15
1 hr.	2	3.0	1.0	2.72	7.30
2	2	3.0	0.7	2.36	7.62
3	2	3.0	0.4	2.52	7.84
4	2	3.0	0.3	1.14	7.50
6	2	3.0	0.2	0.96	7.50
12	2	3.0	0.1	1.06	7.72
18	2	3.0	0.0	----	7.69
24	2	3.0	0.0	0.88	7.70
1 min.	3	3.0	3.0	----	----
5	3	3.0	2.4	2.96	7.16
15	3	3.0	1.8	2.72	7.20
30	3	3.0	1.37	3.68	7.40
1 hr.	3	3.0	0.75	3.04	7.44
2 hr.	3	3.0	0.35	2.64	7.60
3	3	3.0	0.1	2.48	7.84
4	3	3.0	0.0	----	7.50
6	3	3.0	0.0	----	7.60
12	3	3.0	0.0	1.04	7.83
18	3	3.0	0.0	----	7.72
24	3	3.0	0.0	0.84	7.71
1 min.	5	3.0	2.6	----	----
5	5	3.0	1.8	3.76	7.18
15	5	3.0	1.3	3.76	7.50
30	5	3.0	0.67	4.08	7.52
1 hr.	5	3.0	0.5	3.20	7.57
2	5	3.0	0.12	2.88	7.64
3	5	3.0	0.05	2.96	7.69
4	5	3.0	0.0	----	7.91
6	5	3.0	0.0	----	7.70
12	5	3.0	0.0	1.00	7.88
18	5	3.0	0.0	----	7.94
24	5	3.0	0.0	0.88	7.85

FIG. 6.


EFFECT OF TIME
OF CONTACT OF BONE & WATER
IN THE REMOVAL OF
FLUORINE.
(500 CC. OF SOLUTION.)
ORIGINAL F CONTENT 3.0 P.P.M.
2 GMS. BONE
3 GMS. BONE
5 GMS. BONE
FINAL FLUORINE CONTENT (PPM)
3
2
1
0
5
10
15
20
25
TIME IN HOURS.

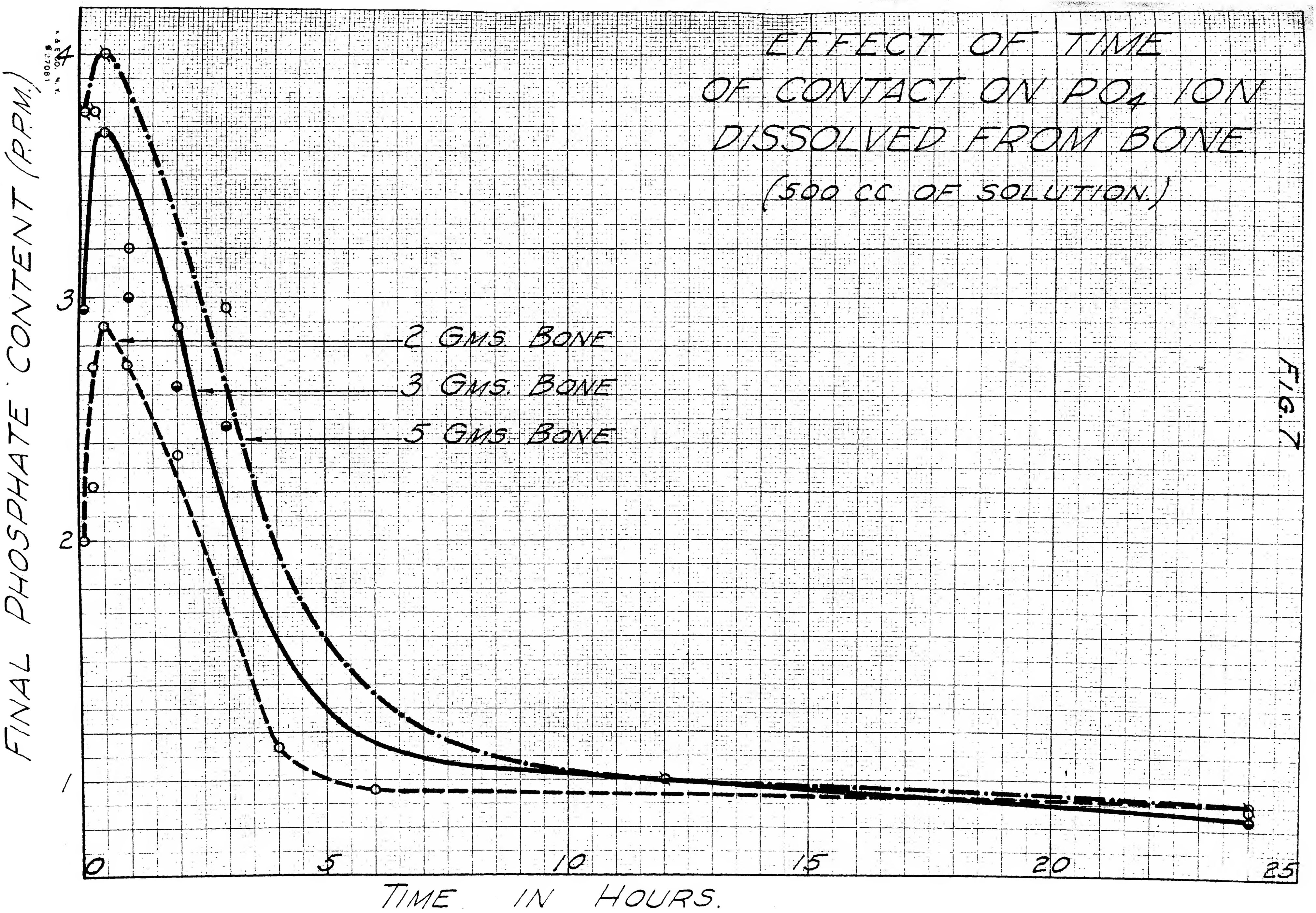
Effect of time of contact on fluorine removal:

Since the rate at which any filter removes fluorine from water is of extreme importance, it became necessary to study the effect of time of contact of bone and water in the removal of fluorine. Instantaneous removal, of course, would be ideal for under this condition the flow of water through the filter bed could be increased to any desired rate. The time of contact, however, was found to play an important part.

Samples of alkali-acid activated bone (20-40 mesh) weighing two, three and five grams respectively, were shaken for one hour in an end-over-end shaker with a solution containing 3 parts per million of fluorine. The mixtures were immediately filtered and the residual fluoride content determined in each case by the Sanchis method. The amount of phosphate in the filtrate was also determined colorimetrically, using ammonium molybdate and stannous tin. Experimental results are shown in Table 19, and Fig. 6.

It will be noted from the graph (Fig. 6) that a family of curves was obtained much as might have been expected. Complete removal is obtained in each case although considerable time is required for the two gram sample of bone to effect the removal. The toxic concentration of fluorine which will cause mottled enamel has been set at 0.9 part per million fluorine (55). Results show that the bone will reduce the concentration to below this toxic level but that the time of contact is important. This slowness of removal brings to light a fundamental consideration in the use of the bone filter; namely, that the capacity of this material is a function of the time of contact, and that the time between regenerations of the bone filter will be increased as the flow rate is decreased. This fact was well brought out in the effect of mesh study (Table 16) where it was found that the longer column of bone removed the greater amount of fluorine.

In making the above equilibrium studies a number of the samples was analyzed for their phosphate largely as a matter of control since it has been shown that the presence of more than 5 to 10 parts per million of this ion interferes with the accuracy of the Sanchis method. The amount of phosphate dissolved is plotted against the time of shaking (Fig. 7). It will be seen from the graph that there is a rapid initial rise in the phosphate concentration and then a rapid decline, the three weights of bone used giving rise to practically the same final phosphate content. Why more phosphate did not go into solution as the time of contact of the bone and the water increased is difficult to explain. It was thought that perhaps a study of the solubility of the bone with respect to phosphate ion in distilled water and carbon-dioxide free water might give some clue. Consequently the above experiment was repeated, three grams of bone being used. Results of this study are shown in Table 20.



EFFECT OF TIME OF CONTACT ON PO_4 ION DISSOLVED FROM BONE

(500 CC. OF SOLUTION.)

FIG. 7

FIG. 8

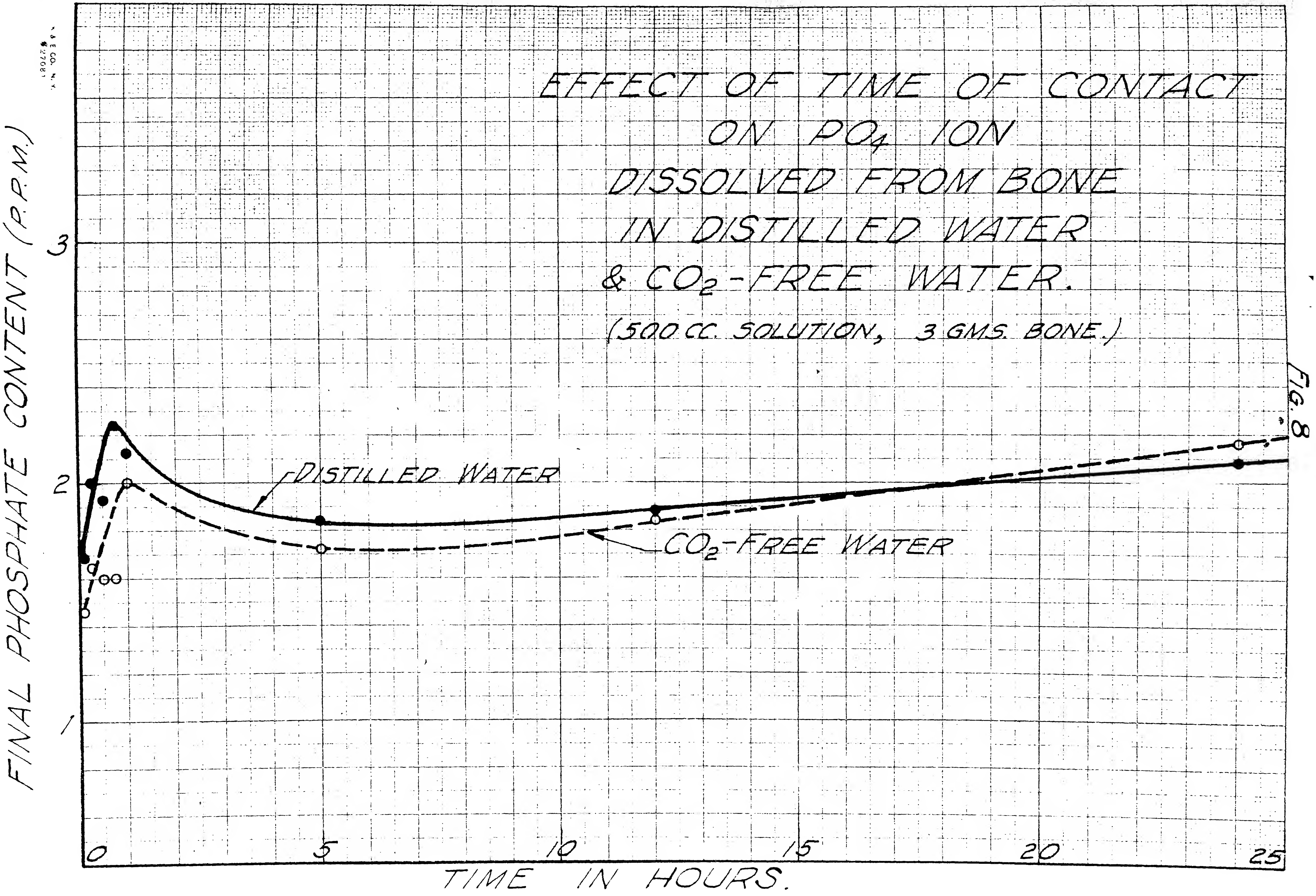


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TABLE 20. EFFECT OF TIME OF CONTACT ON PHOSPHATE DISSOLVED FROM BONE BY DISTILLED WATER AND CARBON - DIOXIDE FREE WATER
(20 - 40 MESH BONE)

Solvent	Time of Shaking	Weight of Bone (Grams)	Phosphate Content (p.p.m.)
Ordinary distilled water	5 min.	3	1.68
	15	3	2.00
	30	3	1.92
	45	3	2.24
	1 hr.	3	2.12
	5	3	1.84
	12	3	1.88
	24	3	2.08
Boiled distilled water (CO_2) free	5 min.	3	1.46
	15	3	1.64
	30	3	1.60
	45	3	1.60
	1 hr.	3	2.00
	5	3	1.72
	12	3	1.84
	24	3	2.16

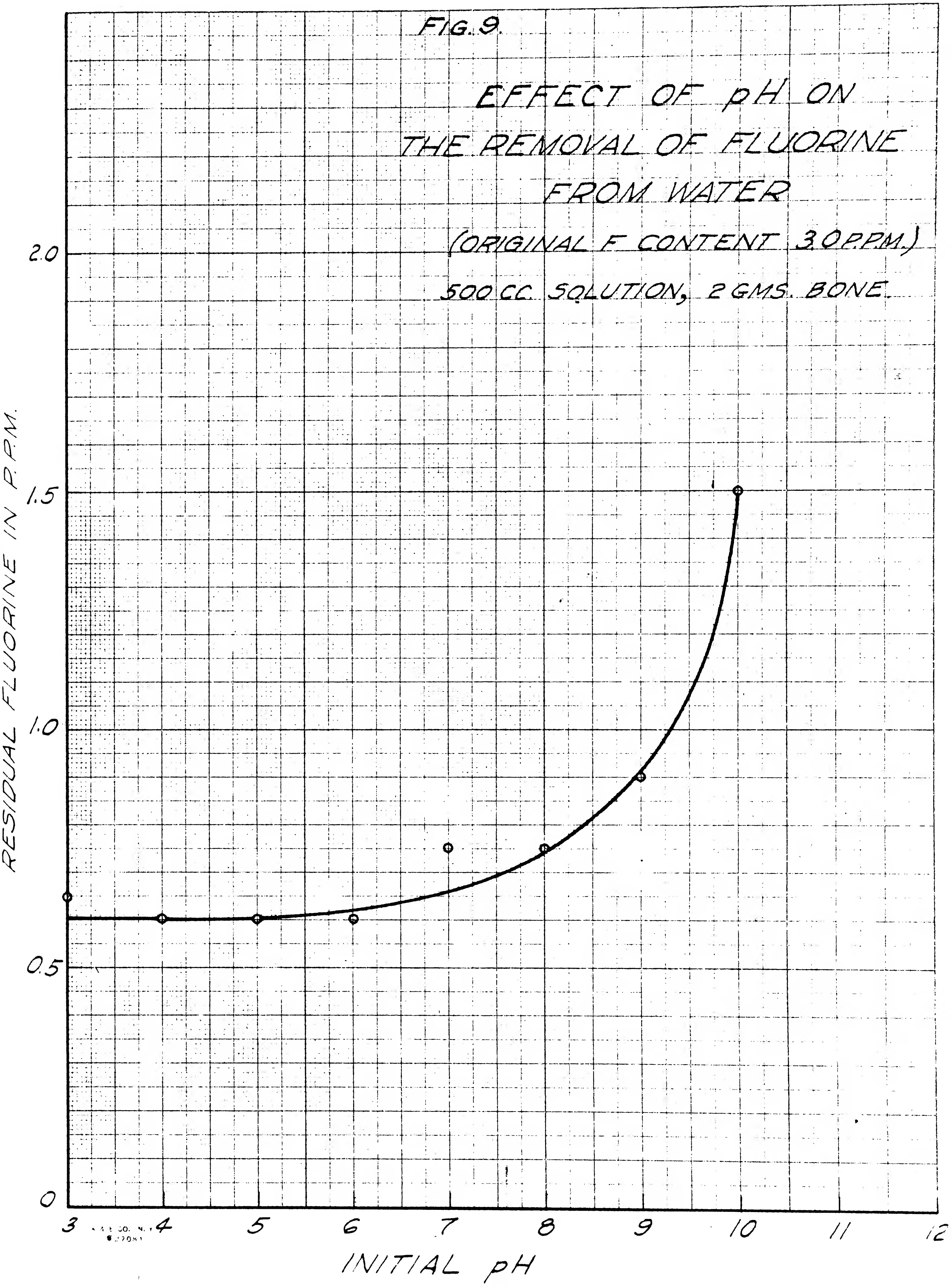
It will be noted that the same rise in solubility takes place, then a decline and the final phosphate concentration approaches the same final value just as in the case of the waters containing fluorine. The total amount of phosphate dissolved, however, was not so great probably due to the fact that a different bone was used in this second study. This "rise and decline" effect may possibly be explained on the basis of a limiting solubility of the phosphate.

FIG. 9.

EFFECT OF pH ON THE REMOVAL OF FLUORINE FROM WATER

(ORIGINAL F CONTENT 3.0 P.P.M.)

500 CC. SOLUTION, 2 GMS. BONE.



Effect of pH on fluorine removal:

Since in so many of the proposed methods of removing fluorine from water the pH played such an important part, it was decided to study this effect in regard to the use of bone. To a series of liter bottles were added 500 cc. portions of a 3 part-per-million fluorine solution. These solutions were then adjusted to different pH values ranging from 3 to 10 by use of the Beckmann pH Meter and adding dilute hydrochloric acid and sodium hydroxide. The contents of each bottle were then shaken with two-gram samples of bone in an end-over-end shaker for one hour, the mixtures immediately filtered and the residual fluorine content of the filtrate determined by the Sanchis method. The final pH was also determined. Results are shown in Table 21 and graphically in Fig. 9.

TABLE 21. EFFECT OF pH ON THE REMOVAL OF FLUORINE FROM WATER

Weight of Bone (grams)	Initial pH	Final pH	Initital F p.p.m.	Residual F p.p.m.
2	3	6.46	3.0	0.65
2	4	6.64	3.0	0.60
2	5	6.93	3.0	0.60
2	6	7.30	3.0	0.60
2	7	7.80	3.0	0.75
2	8	7.95	3.0	0.75
2	9	8.47	3.0	0.90
2	10	10.07	3.0	1.50

It is seen that the pH does have an effect on the ability of bone to remove fluorine from water, the amount removed decreasing with increase in pH. However, no marked decrease is noted till a pH of 9 or higher is reached. Few domestic waters ever attain any such degree of alkalinity, hence the effect is considered unimportant. A rather constant

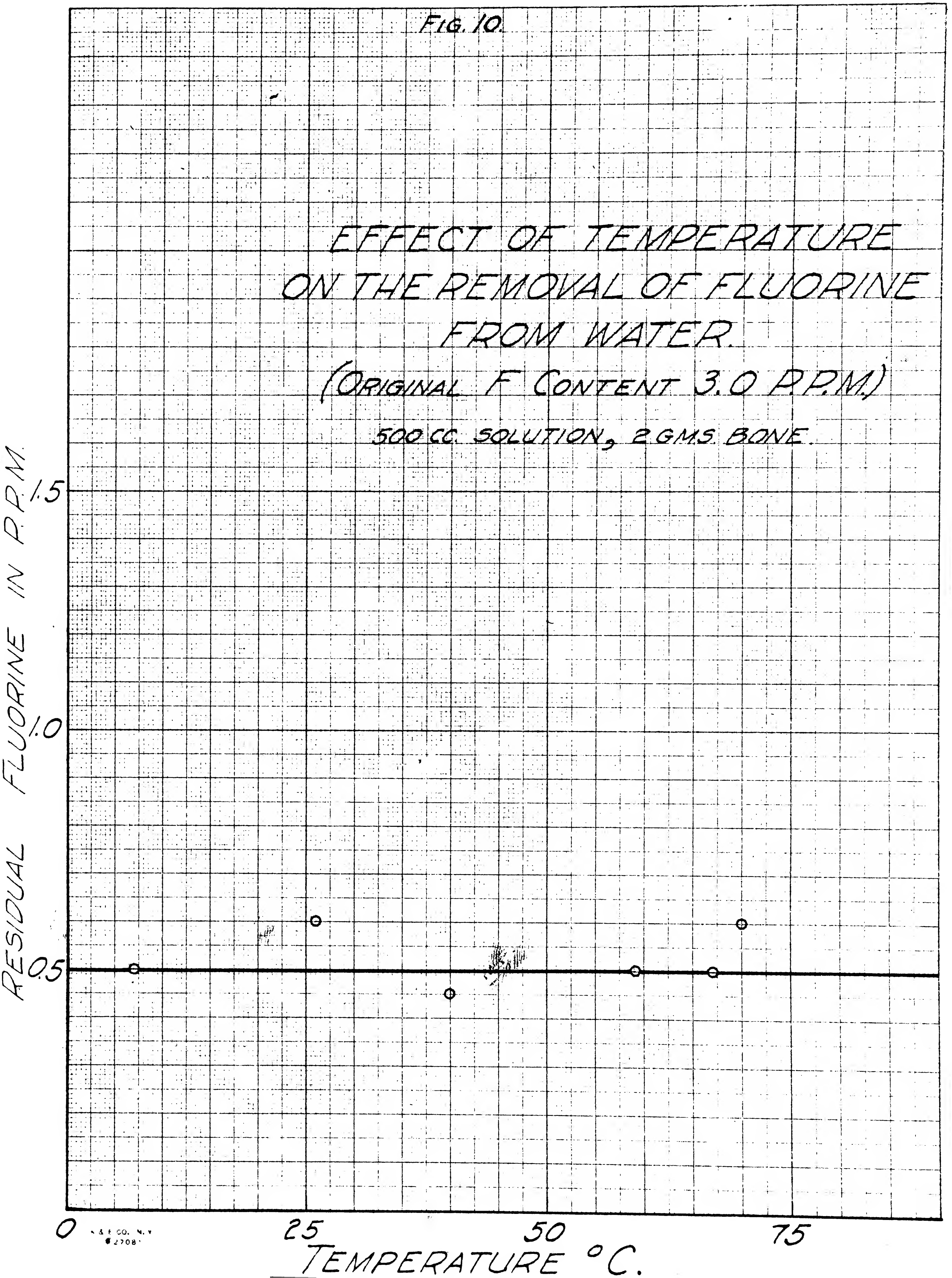


Fig. 10.

EFFECT OF TEMPERATURE ON THE REMOVAL OF FLUORINE FROM WATER.

(ORIGINAL F CONTENT 3.0 P.P.M.)

500 CC. SOLUTION, 2 GMS. BONE.

removal is obtained up to a pH of about 8, the first change beginning to show around neutrality.

The final pH values appear to be significant in that they show that bone has a tendency to bring the reaction mixture to a rather definite value. The average pH value of bone shaken for two hours with a 3 part per million solution is around 7.6. It is noted that there is a tendency for the final pH values of Table 21 to approach this value.

The pH determinations included in Table 19 (effect of time of contact) are likewise significant. The pH values in this case are seen to increase in general with the time of contact. It has been suggested that perhaps this increase may be due to carbonate ion or hydroxyl ion which is replaced somewhat by the fluorine of the water. Assuming that bone is a mixture of carbonate-apatite and hydroxy-apatite, it is not improbable that such a reaction as stated above may be taking place on the surface of the exposed bone.

Effect of temperature on fluorine removal:

Since many reactions are known to take place with greater rapidity at higher temperatures, it was decided to investigate the removal of fluorine at different temperatures. For this purpose several quart thermos jars were used. Standard potency tests with two-gram portions of bone were conducted in these flasks at different temperatures. Results of the experiment (see Table 22, Fig. 10) indicate that there is no effect.

TABLE 22. EFFECT OF TEMPERATURE ON FLUORINE REMOVAL FROM WATER BY BONE. (40 - 60 MESH BONE)

*Temperature °C	Weight of Bone (Grams)	Initial F (p.p.m.)	Residual F (p.p.m.)
7	2	3.0	0.50
26	2	3.0	0.60
40	2	3.0	0.45
42	2	3.0	0.60
59	2	3.0	0.50
62	2	3.0	0.50
67	2	3.0	0.50
67	2	3.0	0.50
70	2	3.0	0.60

*The temperature variation after shaking was less than 1 degree in each case

The residual fluorine in each case is practically of the same order of magnitude, the greatest difference being only 0.15 parts per million of fluorine. This variation is not considered significant since the accuracy of the Sanchis method is only 0.1 part per million. Also the variability of the bone used may account for the difference in some measure.

Effect of salts on fluorine removal:

Since it was thought that the mechanism by which bone removes fluorine from water might involve anion-exchange, the effect of other salts on the removal was investigated to determine if bone is preferential in its "adsorption" of fluorine to other anions. The first study on this subject consisted of the addition of various concentrations of chloride and sulfate ion to 500 cc. of a solution containing 3 parts per million of fluorine. One-andtwo-gram samples of bone were then shaken for one hour with this mixture and the residual fluorine content determined in each case by the Sanchis method. Results of the experiment are given in Table 23. Some slight effect is noted in most cases, but if the correction for sulfate ion is applied to the method, the difference observed is only 0.2 to 0.3 per million. The chloride ion appears to have no effect, the greatest variation being only 0.1 part per million which is within the accuracy of the method. The adsorption seems to be preferential, only a slight effect being found with these two salts commonly found in natural waters.

TABLE 23. EFFECT OF SULFATE AND CHLORIDE ON FLUORINE REMOVAL.

Weight of bone (grams)	Initial Fluorine (p.p.m.)	*Chloride Added (p.p.m.)	**Sulfate Added (P.p.m.)	Final Fluorine (p.p.m.)
1	3.0	0	0	1.75
1	3.0	500	0	1.80
1	3.0	500	500	2.10
1	3.0	0	500	2.10
2	3.0	0	0	1.10
2	3.0	500	0	1.20
2	3.0	500	500	1.50
2	3.0	0	500	1.60

* Chloride added as NaCl

** Sulfate added as Na_2SO_4

The second study conducted on this subject was of a more practical nature. Several representative waters of the State of Arizona as well as one water from Illinois were analyzed for their salt content including fluorine. The results of these analyses are given in Table 24.

TABLE 24. ANALYSIS OF NATURAL WATERS USED IN "SALT-EFFECT" STUDY

	Water	Ca	Mg	CO_3	HCO_3	Cl	SO4	F
1a.	Nine miles West Hayden Junction	22.5	0.0	62.4	578.3	662.	520	3.6
2a.	Tacna, Ariz.	97.5	127.5	42.0	187.8	681.	2700.	3.5
3a.	Aledo, Ill.	45.0	18.7	24.0	173.2	214.	900.	2.5
4a.	Agua Caliente Hot Springs	7.5	11.2	14.4	102.4	273	200.	5.0
5a.	Mammoth (Mine)	22.5	0.0	19.2	153.7	49	40.	2.8

All results are expressed as parts per million
Corrections applied for sulfate ion in each case

Standard fluorine solutions made up in distilled water were prepared, each solution corresponding in fluoride content to one of the natural waters. To each of the two corresponding water samples (the natural water and the standard) was added an equal amount of bone, the mixtures shaken side-by-side for one hour in an end-over-end shaker, filtered, and the residual fluorine content determined by the Sanchis method. Results of the experiment are given in Table 25.

to become mottled later. Deciduous or baby teeth more rarely show mottling, being chiefly a defect of the permanent teeth, although a few cases in the temporary molars among the Indian children in Arizona have been observed (62). Concentrations over 10 parts per million may produce mottled enamel in deciduous or baby teeth (59).

Mottled enamel has been produced experimentally in albino rats, in guinea pigs, and in dogs by the feeding of fluorides or the residue of water from a community which contains fluorides (61). A number of lines of evidence indicate that the fluorine passes into the blood stream and interferes with the calcification of the unerupted teeth of children. It does not act in the mouth upon the enamel of the erupted portion of the teeth. The teeth of children or adults who do not begin drinking water containing fluorine until after their second sets of teeth have erupted do not later show mottled enamel. In humans, the enamel organ disappears as soon as the enamel of the permanent teeth is calcified. The enamel does not regenerate itself and behaves, therefore, like dead tissue. For this reason the enamel of the teeth of adults is unaffected by the drinking of water containing fluorine (60).

It is obvious that mottled teeth are very disfiguring and ugly, for the stain which commonly filters into the porous areas of the teeth is usually rust colored. In addition, mottled teeth are defective in formation and calcification and are, therefore, structurally weak. The defect is irreparable and permanent. It has been estimated by the Tucson Dental Association that it would cost $1,000 for dental care of the teeth of the average person with mottled enamel up to adulthood, at which time the teeth must usually be replaced by false ones (60).

TABLE 25. REMOVAL OF FLUORINE FROM NATURAL WATERS AND STANDARD FLUORINE SOLUTIONS OF THE SAME CONCENTRATION

Water	Initial F (p.p.m.)	Weight of bone (Grams)	Residual F (p.p.m.)
1a	3.6	2	1.30
b	3.6	2	1.25
2a	3.5	2	1.35
b	3.5	2	1.25
3a	2.5	2	1.10
b	2.5	2	0.80
4a	5.0	3	1.50
b	5.0	3	1.40
5a	2.8	2	1.10
b	2.8	2	0.80

a = natural water
b = corresponding standard fluorine solution

It will be noted that the fluoride removing power of the bone is slightly decreased in the natural waters, indicating that the "adsorption" of fluorine by bone is preferential; or, if not preferential, the presence of other ions in the solution does not have any appreciable effect.

Effect of double treatment on fluorine removal:

It was suggested that a double treatment of water with smaller portions of bone might prove more effective in fluoride removal than a single treatment with twice as much bone. The double treatment procedure has been found to function well in the extraction of gold by means of activated carbon, and, hence it was tried on fluorine removal by bone. Successive treatments of a standard fluorine solution with two 1-gram samples were compared with results obtained using two gram samples. In the case of the double treatment, 500 cc. of a 3 part-per-million fluorine solution was shaken for one hour on an end-over-end shaker with a one-gram sample of bone, the mixture immediately filtered, and a second one-gram portion of bone added to the filtrate and shaken again for one hour. For purposes of comparison two other 500 cc. volumes of standard fluorine solution were shaken with two-gram samples of bone for one hour and two hours respectively. Residual fluorine in each case was determined by filtering the mixtures and analyzing the filtrate.

The results are shown in Table 26.

TABLE 26. EFFECT OF DOUBLE-TREATMENT OF WATER WITH BONE:

Treatment	Initial F (p.p.m.)	Residual F (p.p.m.)
(1) 1 gram bone shaken with 500 cc. of solution for one hour, filtered, and a second fresh 1 gram portion added and shaken for one hour.	3.0	0.7
(2) 2 grams bone shaken for 1 hour	3.0	0.8
(3) 2 grams bone shaken for 2 hours	3.0	0.75

Results indicate that the double treatment is no more effective than the single treatment with twice the quantity of bone being used. It is conceivable, however, that under conditions where a water is high in fluorine and a considerable demand for water exists, a double pressure-filter-treatment might prove effective in removing greater amounts of the fluorine. For ordinary use the single unit has been found to function satisfactorily.

The thin-section photographs which are presented were taken using the following specifications:

Magnification 105 X

#3 Leitz objective

#2 Leitz eye piece

B (green) filter

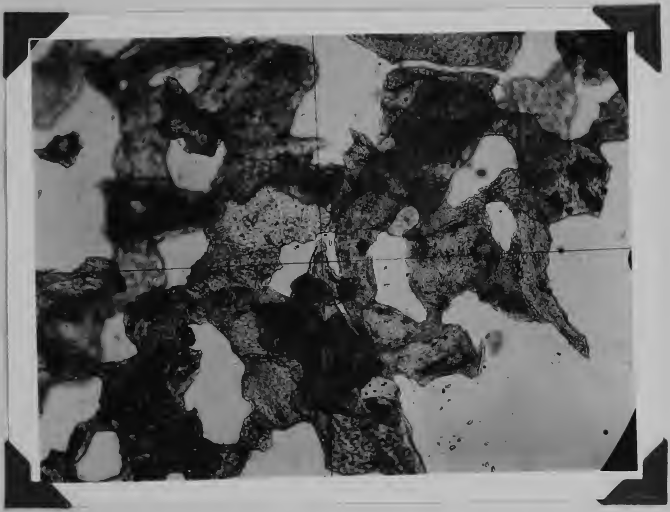


Fig. 10. Steamed bone meal, 40 - 60 mesh, original untreated sample

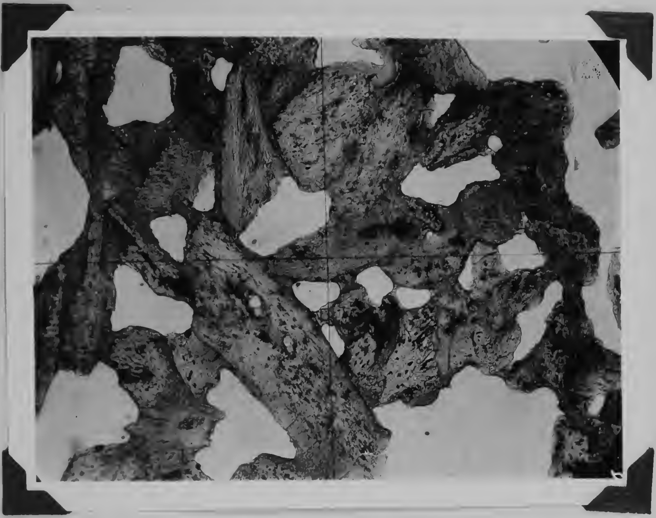


Fig. 11. Steamed bone meal, 40 - 60 mesh, alkali-acid treated

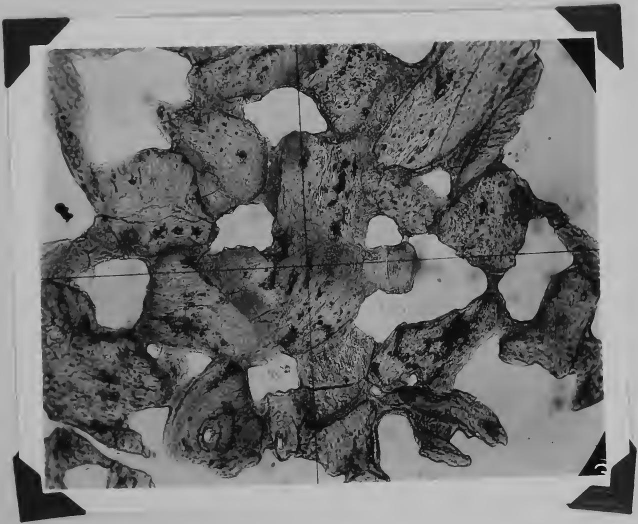


Fig. 12. Steamed bone meal, 40 - 60 mesh, alkali-acid treated, fluorine saturated

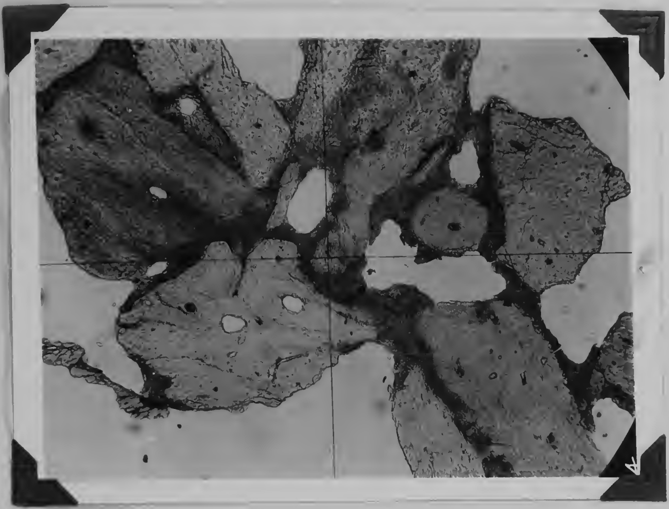


Fig. 13. Steamed bone meal, 20 - 40 mesh, alkali-acid treated, calcined, and acid treated

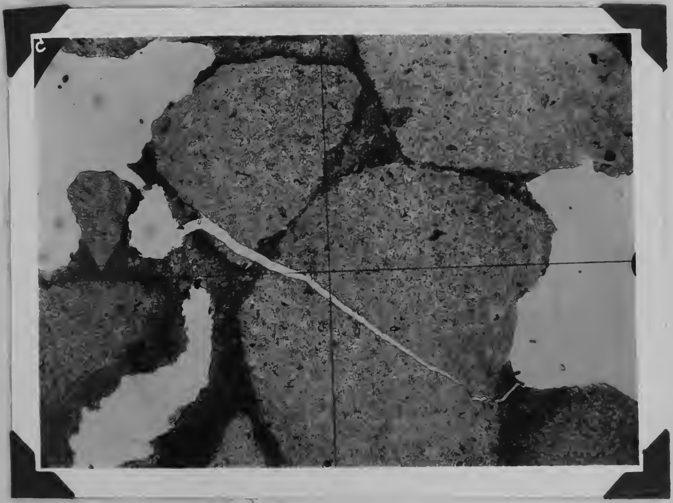


Fig. 14. "Defluorite", synthetic tri-calcium phosphate, 20 - 40 mesh

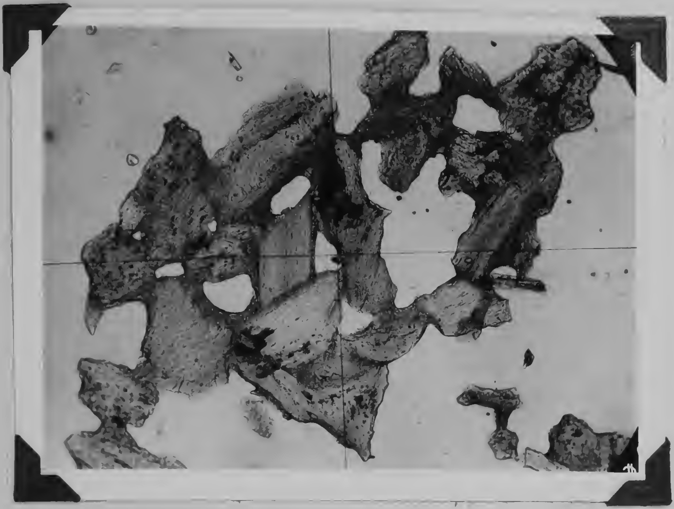


Fig. 15. Steamed bone meal, 40 - 60 mesh, alkali-carbon dioxide treated

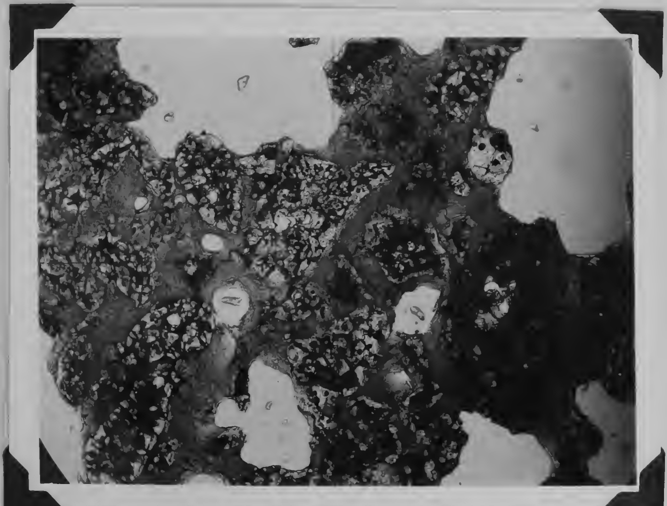


Fig. 16. Rock phosphate, untreated

Photomicrographs of bone:

It seemed advisable to gain some knowledge of the structure of bone in connection with the removal of fluorine from water. Consequently, several thin sections of bone were prepared and photographed along with a sample of "Defluorite" and rock phosphate. The photographs are shown in Figs. 10 to 16 inclusive.

The porosity of the bone and "Defluorite" is evident from the pictures, perhaps explaining in some measure the greater fluoride removing power of these two materials over that of the rock phosphate.

Mechanism of the reaction:

When the work on bone was first begun in 1934 at the University of Arizona by H. V. Smith, the formula of bone was considered to be $(Ca_3(PO_4)_2)n{\cdot}CaCO_3$ where n was 2 or 3. It was assumed that the sodium hydroxide treatment resulted first in the formation of a hydroxy apatite; that the following hydrochloric acid formed the chlorapatite; and finally that the fluorine of the water displaced the chloride radical with the formation of a fluorapatite thus removing the fluorine from the water. The possibility of anion-exchange was first suggested by Gassmann (27). The following series of equations may help to show the supposed reactions.

(1) $(Ca_3(PO_4)_2)n{\cdot}CaCO_3 + 2NaOH \longrightarrow (Ca_3(PO_4)_2)n{\cdot}Ca(OH)_2 + Na_2CO_3$

(2) $(Ca_3(PO_4)_2)n{\cdot}Ca(OH)_2 + 2HCl \longrightarrow (Ca_3(PO_4)_2)n{\cdot}CaCl_2 + 2H_2O$

(3) $(Ca_3(PO_4)_2)n{\cdot}CaCl_2 + NaF \longrightarrow (Ca_3(PO_4)_2)n{\cdot}CaF_2 + 2NaCl$

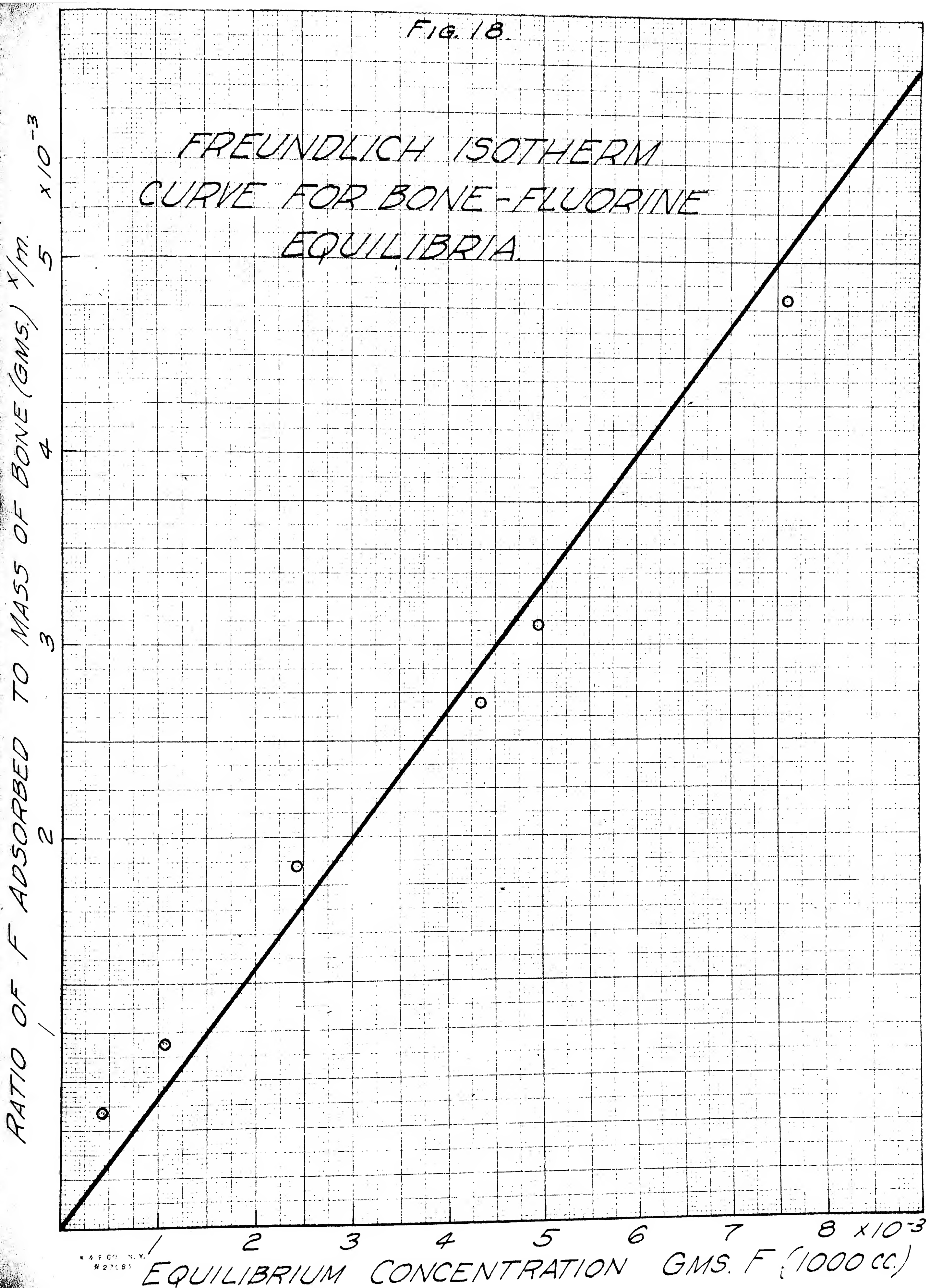
This assumption may not be true for several reasons; namely (1) the amount of fluorine taken up by the bone from the water does not approach the theoretical value assuming a complete conversion of the chlor-apatite to the fluor-apatite, but since the reaction may only be a surface phenomenon, this reason may not be important, (2) the chloride content of the bone after treatment with HCl has been found by analysis to be less than 0.10% chloride, indicating that the second step is only partial; (3) assuming the reaction to anion-exchange, it should be possible to regenerate the bone with high concentrations of NaCl, but this has been found impossible; (4) regeneration of the bone can be accomplished with a number of acids, including organic acids, which indicates that the formation of a chlor-apatite is not necessary for the success of the removal, but that the acid serves possibly only to neutralize the excess alkalinity; and (5) no evidence of a true chemical reaction (anion-exchange exclusively) is found when an adsorption isotherm is determined for the bone and the fluorine.

The true mechanism of the reaction is still in a conjectural stage. An adsorption isotherm for the reaction was determined by use of the well-known Freundlich equation. Results of this experiment indicate that the mechanism is neither true chemical reaction nor is it adsorption alone. The Freundlich equation is as follows:

$$x/m = KC^{1/p}$$

where x is the grams of material adsorbed (fluorine)
m is the grams of adsorbing material (bone)
C is the equilibrium concentration with respect to the adsorbed ion (fluorine)
K and $1/p$ are constants for each particular system.

FIG. 18.



FREUNDLICH ISOTHERM CURVE FOR BONE-FLUORINE EQUILIBRIA.

The details of the study were as follows: Samples of activated bone, weighing two grams, were shaken several days (equilibrium) in glass stoppered bottles with fluorine solutions ranging in concentration from 0 to 30 parts per million; the mixtures were filtered, and the residual fluorine of the solution (C) determined in each case by titration with standard thorium nitrate solution using the Rowley and Churchill modification of the Willard and Winter method. The ratio x/m was plotted against C (see Fig. 18). According to Freundlich, the curve for a true adsorption process is exponential, and for a simple chemical reaction the curve obtained parallels the "C" axis. However, the curve obtained in this study is seen to be a straight line of positive slope indicating the formation of a solution of varying composition, which in this case would obviously be solid-solution formation. Other investigators have also noted this tendency toward solid-solution formation. Hendricks, Hill, Jacob, and Jefferson (29) in discussing the composition of bone and phosphate rock states that "it would be expected that carbonate apatite (bone) could form solid solutions with hydroxy fluor-,oxy-, or a possible sulfate apatite. Many of the minerals mentioned in the first part of the article are members of such solid solutions". This then might possibly be the mechanism. It is conceivable that the treatment of the bone with sodium hydroxide might cause a partial conversion to the hydroxy-apatite. The hydroxy-apatite thus formed might react with fluorine of the water to form a certain amount of hydroxy-fluor-apatite, which, according to Hendricks, might be expected to form solid solutions with carbonate apatite (bone). Removal of fluorides from water would thus be effected. The acid treatment would serve merely to neutralize the

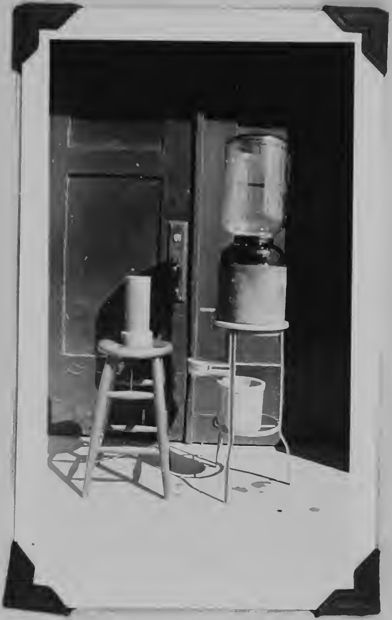


Fig. 19. "Olla" type cooler drinking fountain showing cartrige at left.

excess alkali, since it has been shown that the removal takes place best at a pH below 8. That any displacement which occurs involves the hydroxyl or carbonate group rather than the chloride radical is substantiated by the pH measurements shown in Table 19. It is noted that the pH of the reacting mixture increases with time of contact of the bone with the water. Since the amount of fluorine removal increases with the time of contact, is is very probable that the increase in pH noted is due to the partial displacement of either the hydroxyl or carbonate group. After this partial reaction solid-solutions may be formed.

Practical applications:

The practicability of the use of bone for removing fluorine from drinking water has been demonstrated by a number of laboratory and field studies. Two types of filter have been designed for home and school use --the "olla" filter and the "pressure" filter.

The "olla" filter consists of an unglazed earthen-jar (olla cooler) drinking fountain (see Fig. 19). These coolers are common equipment in offices and homes. A charge of bone is put into a 3 x 11 inch cylinder which is then closed top and bottom by means of fine mesh brass screens. The cylinder is then placed in the olla and a 5 gallon bottle of water set in place on top of the cooler. The apparatus is so designed that the neck of the bottle fits an inch or two into the cylinder in such a way that the water must filter down through the bone and out the bottom of the cylinder. Percolation continues until the water level outside in the olla becomes the same as that in the cylinder. Water may then be drawn off at will from the tap. When water is removed by means of the tap,more water from the bottle percolates down through the bone in the cylinder until equilibrium is again established between the two water levels. In this manner a reserve supply of treated water becomes available for use.

Several studies were conducted on this type of filter with good results. The first of such a study involved the use of a cartridge of bone containing 1.59 pounds of bone. The cylinder in this case consisted of 3 x 11 inch transit pipe which was filled to a depth of about 9 inches with 40-60 mesh bone and then sealed in by means of the brass screens. Water containing 3.5 parts per million of fluorine was passed through the bone as rapidly as possible. Samples were taken after the passage of every 5 gallons and analyzed for fluorine by the Sanchis method. A total volume of 165 gallons of the water was defluorinated by the 1.59 pounds of bone before the toxic level of 0.9 of a part per million of fluorine was reached. This corresponds to the removal of 1.37 grams of fluorine per pound of bone or 67.13 gms. per cubic foot of bone. Results of the study are shown in Table 27.

TABLE 27. REMOVAL OF FLUORINE WITH THE OLLA TYPE FILTER (40 - 60 MESH BONE)

Weight of Bone in Cartridge (pounds)	Volume of Water Treated (gallons)		Fluorine Content (p.p.m.)
1.590	0	(control)	3.50
	50		0.00
	100		0.00
	150		0.00
	155		0.10
	160		0.35
	165		0.75
	170		1.20
1.799	0	(control)	11.00
	25		0.00
	50		0.00
	55		0.00
	60		0.50
	65		1.50

The second study involving the olla filter consisted in the use of a cartridge of bone containing 1.799 pounds of bone through which was passed a water containing 11 parts per million of fluorine. This cartridge of bone treated 60 gallons of water which corresponds to a removal of 1.43 grams of fluorine per pound of bone or 70.1 grams per cubic foot of bone. It is of interest to note that the total amount of fluorine removed per cubic foot of bone is the same regardless of the initial concentration of the fluorine. Results of this study are also shown in Table 27.

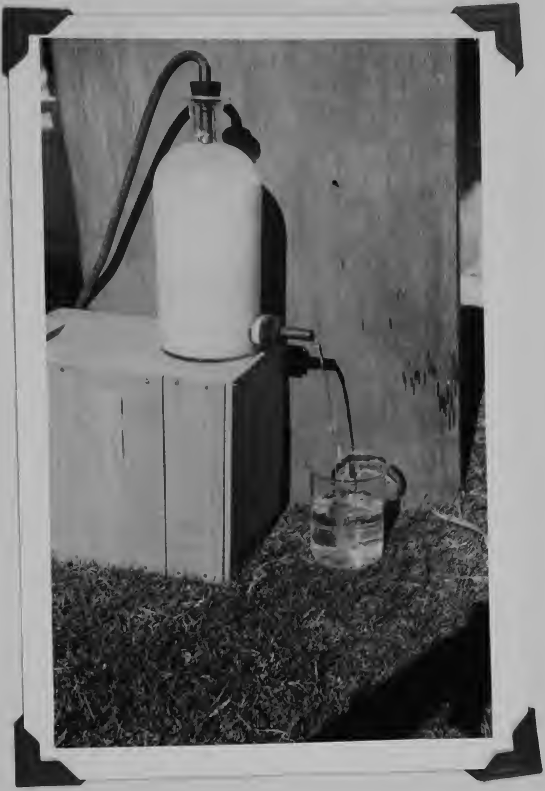


Fig. 20. "Pressure" type filter in action. Later filters have been made of steel since the glass has been found too easily breakable.

The second type of filter designed, namely the pressure filter, is more applicable to those situations in which a large quantity of water is needed in a short time. For general domestic use including cooking and drinking and for use in schools, this type of filter is best adapted. The unit consists of a closed cylinder with an inlet for the water at the top and an outlet at the bottom. The opening is designed to produce a spray effect when the water is passed to prevent channeling. The bottom outlet is covered with a fine mesh screen to prevent loss of the bone. The size of the cylinder depends upon the initial fluoride content of the water and the demands made upon it. In most of the work carried out in the field a 10 to 12 pound unit was used,

A number of field trials have been conducted throughout the state with this pressure type filter. The most extensive, however, were those carried on at the Pima County Preventorium, an institution organized for the purpose of "building-up" children suspected of having tuberculosis or who have been in contact with the disease. The age of the children varies from about eight to fifteen years, just the age at which mottled enamel is most likely to occur. When it was found in May, 1938 that the water used by the children (130 in all) was toxic, containing 4.0 parts per million of fluorine, it was suggested that these bone filters be installed. And since that time these pressure units have been supplied. Daily samples of the effluent water were taken by the attendant at the Preventorium and sent to the laboratory for analysis. In this way a closely controlled series of field experiments have been conducted. Results of one of the 36 trials so far conducted by a pressure unit containing 11 pounds of bone is shown in Table 28.

TABLE 28. REMOVAL OF FLUORINE WITH BONE IN THE PRESSURE FILTER (40 - 60 MESH BONE) (ORACLE PREVENTORIUM)

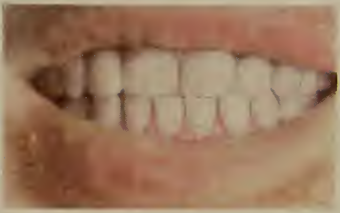
Volume Passed (Gallons)	Fluoride Content (P.p.m.)
Control	4.0
90	0.5
450	0.0
835	0.0
888	0.75
968	0.80

It is seen that the bone removed a total of 14.5 grams of fluorine in treating the 968 gallons of 4.0 part-per-million fluorine water. This corresponds to a removal of 1.32 grams of fluorine per pound of bone or 64.7 grams per cubic foot of bone.

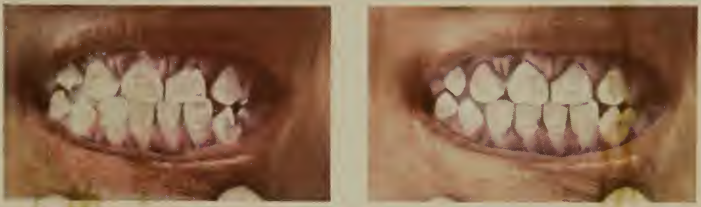
Removal of fluorine from cider:

During recent years it has become the practice to spray fruits and vegetables with fluorine sprays. This procedure is particularly prevalent in the Pacific Northwest. First-grade fruit can bear the cost of fluorine removal by washing, but the cost of cleaning second grade fruit and culls which are used for making cider is not justified economically. Therefore, much of the cider produced in this region carries high concentrations of fluorine. The removal of this element presents a serious problem to the orchardist. The successful use of bone for the removal of fluorine from drinking water suggested the possibility of using this same treatment on cider containing high concentrations of fluorine.

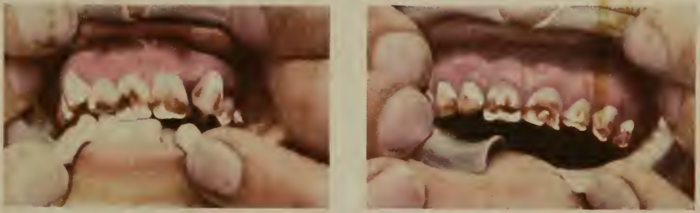
The first study conducted on this problem consisted of the preparation of standard fluorine-cider solutions by adding standard sodium



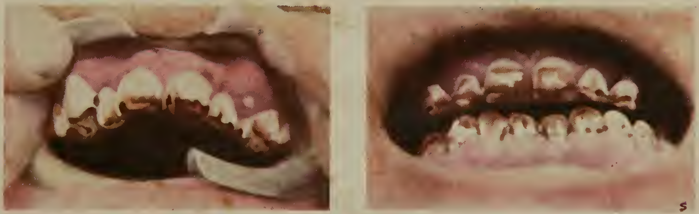
NO MOTTLING



MILD MOTTLING



MODERATELY SEVERE MOTTLING



SEVERE MOTTLING

Types of Mottled Enamel Found in Arizona

Fig. 1. Typical cases of mottled enamel found in certain areas in Arizona (courtesy H. V. and M. C. Smith)

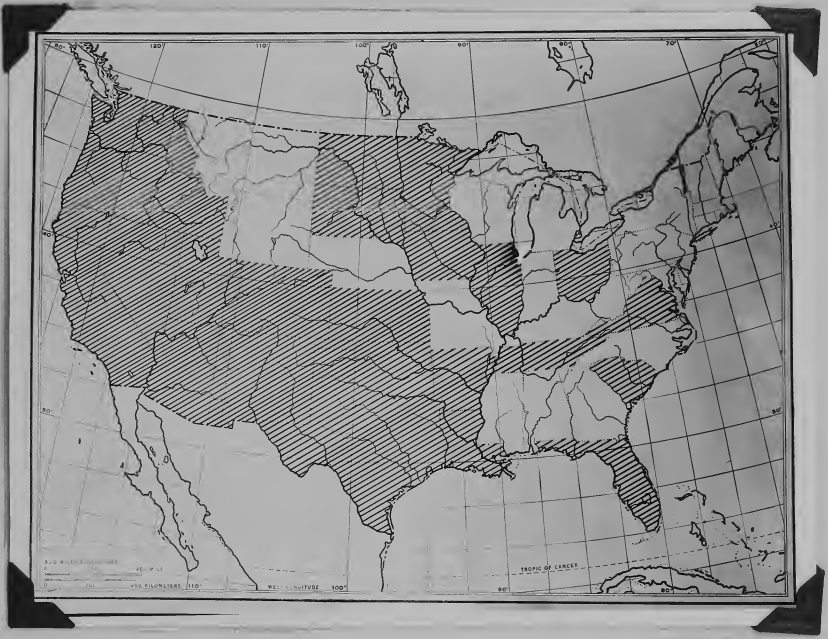


Fig. 2. Distribution of mottled enamel in the United States. States which are cross-hatched have reported cases of mottled enamel. Since this article was written, mottled enamel has been reported in Wisconsin, Indiana, and Mississippi.

The typical cases of mottled enamel found among the native-born inhabitants of certain sections of Arizona are shown in Fig. 1. Mottled enamel of the very mild types is distinguished by dull, opaque, paper-white areas only. The teeth are normal in form and shape but not in color. In the more severe types, a greater proportion of the surface of the tooth is affected and there is an accompanying discoloration, especially noticeable on the teeth most exposed. Mottled enamel which is severe in type is further characterized by pitting and corrosion (61).

This dental defect has been noted over a widespread area. Instances of its occurrence have been reported in England, Holland, Italy, North Africa, Mexico, Spain, China, Japan, several South American countries, and in 24 states in this country. The distribution of mottled enamel in the United States is shown in Fig. 2. Since fluorine is so common in nature, ranking twentieth in quantity in the earth's crust, it is not unnatural that fluorine should find its way into many water supplies. Nine deposits of fluorspar (CaF_2) have been reported in Arizona alone, and other deposits throughout the United States must in all probability contribute to the fluoride content of waters. Waters flowing over other minerals containing fluorine, such as tourmaline, biotite, and apatite, must also dissolve their share of fluorine.

The continuous use of water containing over 0.9 of a part per million of fluorine during the period of calcification of the teeth has been found to cause mottled enamel. This toxic limit was determined by correlating tooth conditions with concentrations of fluorine in many water supplies in Arizona (55).

fluoride to the cider. These solutions were then shaken for one hour in an end-over-end shaker with different amounts of bone and the residual fluorine content then determined. The method of analysis for fluorine becomes quite complex when organic matter is present, such as in the case of cider. Under these conditions a preliminary 16 to 24 hour ashing with especially pure calcium hydroxide is needed. The fluorine content is then determined by distillation of the ash under the Willard and Winter procedure. Results of this study are shown in Table 29.

TABLE 29. REMOVAL OF FLUORINE FROM CIDER BY SHAKING WITH BONE.

		Weight of bone (grams)	F content (p.p.m.)
(1)	Original cider	0	0.72
(2)	242.5 cc. cider plus 7.5 cc. of 100 p.p.m. solution of fluorine--gives approx. 3 p.p.m. F	1	0.37
(3)	247.5 cc. cider plus 2.5 cc. of 1000 p.p.m. solution of F--gives approximately 10 p.p.m. F	5	0.56

It is seen that the cider used did contain some fluorine and that the additional fluorine added was removed by shaking with the bone.

The second study conducted on this problem consisted in passing a 10 part per million fluorine-cider solution through a column of 100 grams of bone in a glass tube whose diameter was 2.2 cms. The bone used was 40-60 mesh calcined bone. Samples were taken at the end of the passage of every 200 cc. and analyzed for the fluoride content by ashing and distilling as explained above. Results are shown in Table 30.

TABLE 30. REMOVAL OF FLUORINE FROM CIDER BY A COLUMN OF BONE (40 - 60 MESH CALCINED)

Sample	Initial F (p.p.m.)	Residual F (p.p.m.)
1 st. 200 cc.	10	0.32
2 nd. 200 cc.	10	0.15
3 rd. 200 cc.	10	0.35
4 th. 200 cc.	10	0.10
5 th. 200 cc.	10	0.28
6 th. 200 cc	10	0.20

Fluorine removal by other phosphates:

A number of materials which were either similar to bone or which it was thought might have possibilities for use in fluorine removal were investigated. Most of these studies involved shaking the material with a standard fluorine solution and noting the residual fluorine content of the filtrate. None of the materials tried showed any marked fluoride removing power with the exception of bone-black, but five times as much of this material was necessary to effect the same removal as did the activated bone. The standard potency test was conducted on these other phosphates using in some cases 10 grams of the material instead of two grams as called for by the test. Results of this study are shown in Table 31.

TABLE 31. FLUORIDE REMOVING POWER OF OTHER PHOSPHATES

Material and Treatment	Weight of Material used (grams)	Residual Fluorine (p.p.m.)
Aluminum phosphate (powder--not treated)	2	1.0
Ferric phosphate (powder--not treated)	2	2.2
Fla. rock phosphate (raw rock--alkali and acid treated)	2	2.6
Fla. rock phosphate (calcined rock--alkali and acid treated)	2	1.75
Jacobs rock phosphate (calcined at 1400 deg.C.)	2	2.3
Jacobs rock phosphate (alkali-acid treated)	10	1.5
Bone black (untreated)	2	1.5
Bone black (alkali-acid treated)	10	0.6
$Ca_3(PO_4)_2$ and $CaCO_3$ mixture, 300 gms. and 50 grams	2	1.25
Powdered egg-shell (alkali-acid treated)	2	1.75
Calcined bone (alkali-acid treated, calcined, acid treated)	2	0.57

Other tests included the precipitation of tri-calcium phosphate from solutions containing fluorine as suggested by Mac Intire (37). This study involves mixing phosphoric acid, calcium hydroxide, and clay and allowing the precipitate formed to settle out. According to Mac Intire good removal should result; but the experiment proved a failure when tried at Arizona. Results of the study are shown in Table 32.

TABLE 32. REMOVAL OF FLUORINE BY PRECIPITATION METHOD

Initial Fluorine (p.p.m.)	Suspended Matter Added (p.p.m.)	Calcium Hydroxide Added (p.p.m.)	Phosphoric Acid Added (p.p.m.)	Residual Fluorine (p.p.m.)
6.0	250	271	190	6.0

Field experiments using "Defluorite" (synthetic tri-calcium phosphate) as furnished by the National Aluminate Corporation of Chicago, Illinois, have also been tried. However, the tests in Arizona have not shown this material to possess the fluoride removing power as claimed by the manufacturers. The field experiments in this case were conducted by Mr. E. O. Foster, at that time Assistant Chemist for the Arizona Agricultural Experiment Station, branch laboratory at Phoenix, Arizona. A 10 pound unit of Defluorite was used and water from the Date Garden at Tempe, Arizona was passed through and sampled periodically. Results of the study are shown in Table 33.

TABLE 33. REMOVAL OF FLUORINE WITH "DEFLUORITE" IN THE PRESSURE FILTER. (TEMPE, ARIZONA)

Volume passed (Gallons)	Fluoride Content (p.p.m.)
Control	1.7
406	0.2
763	0.2
915	0.0
1347	0.2
1703	0.5
1780	0.7
1907	0.8
1932	0.9

Ten pounds of "Defluorite" treated 1907 gallons of water containing 1.7 parts per million of fluorine. This corresponds to a removal of 10.05 grams of fluorine per 10 pounds of the material or 1.005 grams per pound. However, the removal is only 29.1 grams of fluorine per cubic foot of "Defluorite". The quantity of fluorine removed by one cubic foot of bone is thus seen to be more than twice as great as that removed by a cubic foot of "Defluorite". One cubic foot of bone weighs 49 pounds whereas one cubic foot of "Defluorite" weighs only 29 pounds. This is considered to be a point in favor of bone since the volume of bone that can be put into a filter is more important than the weight of bone that can be used.

Another field study using "Defluorite" was conducted at the orchard of Mr. A. C. Presscott of Phoenix, Arizona. The unit tested also contained ten pounds of the material. Results of the experiment are shown in Table 34.

TABLE 34. REMOVAL OF FLUORINE WITH "DEFLUORITE" IN THE PRESSURE FILTER.

Volume of Water Passed (Gallons)	Phosphate Dissolved (p.p.m.)	Fluorine Content (p.p.m.)
0 control	--	8.10
7.5	200	0.18
22.5	130	0.18
45.0	60	0.27
75.0	41	0.27
172.5	14	0.30
267.7	--	0.25
365.7	--	0.50
401.0	--	1.50

FIG. 21.

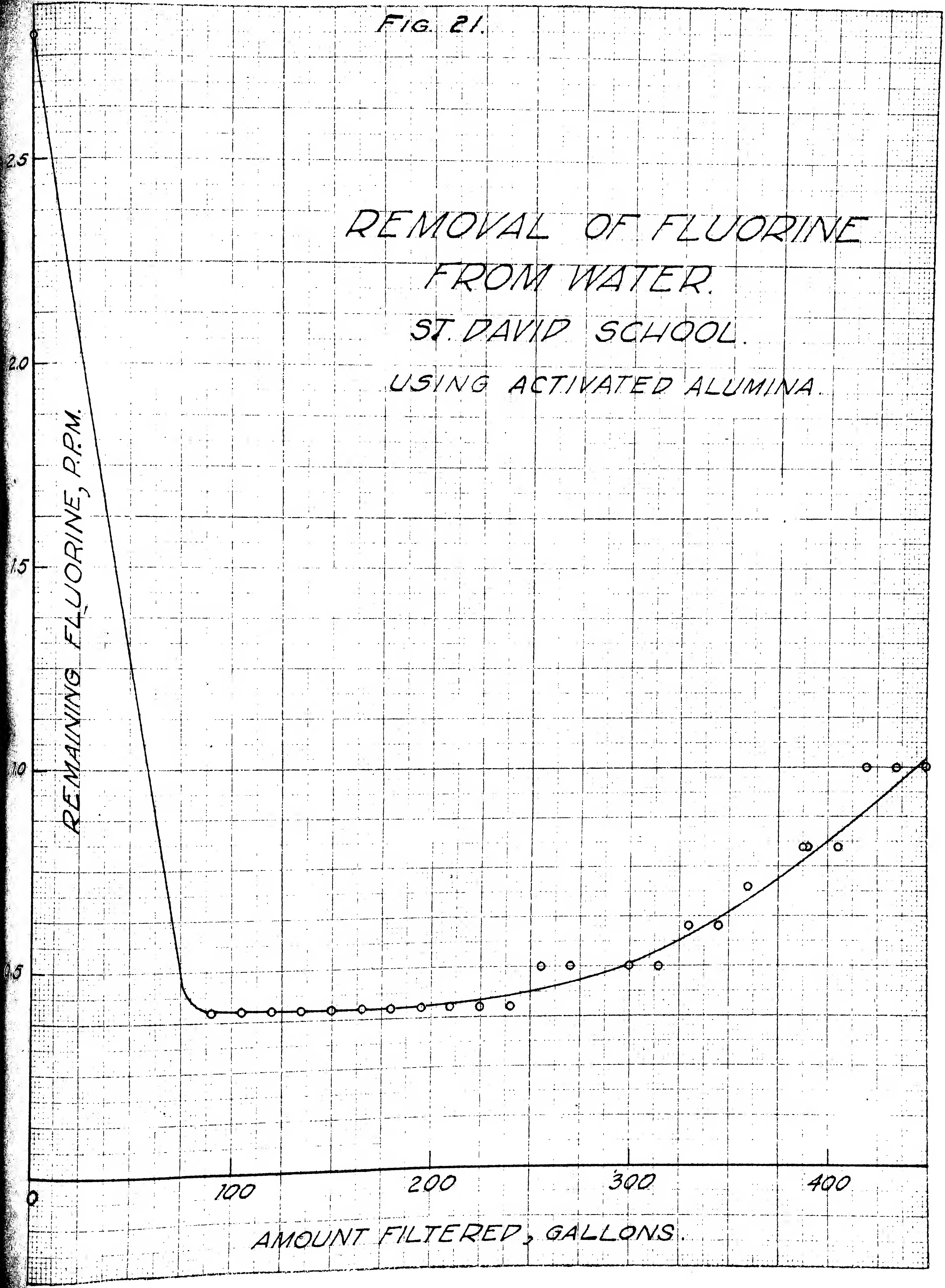
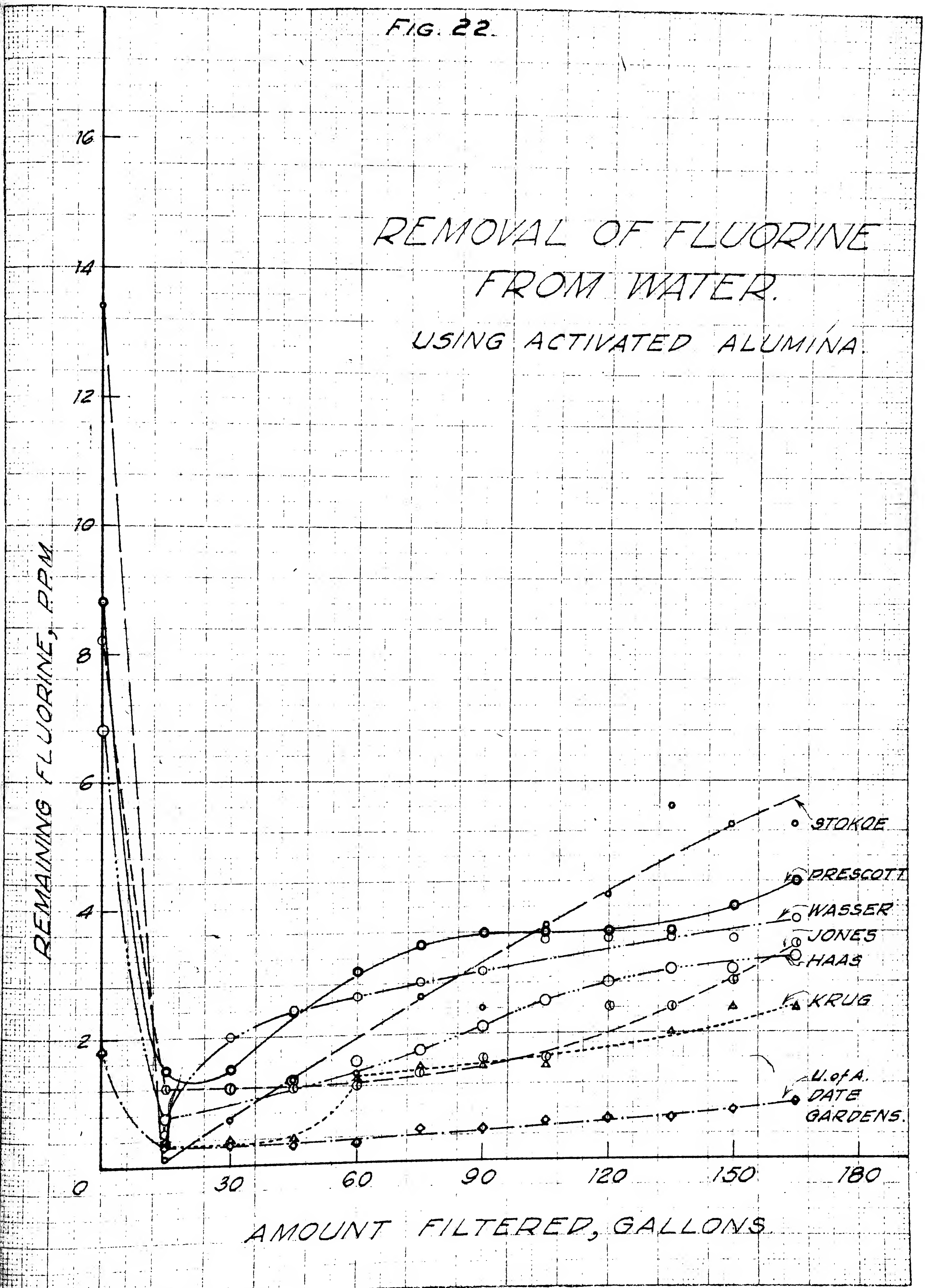

REMOVAL OF FLUORINE FROM WATER.
ST. DAVID SCHOOL.
USING ACTIVATED ALUMINA.
REMAINING FLUORINE, P.P.M.
2.5
2.0
1.5
1.0
0.5
0
100
200
300
400
AMOUNT FILTERED, GALLONS.

FIG. 22.



REMOVAL OF FLUORINE FROM WATER.

USING ACTIVATED ALUMINA.

This unit removed 11.1 grams of fluorine for the ten pounds of material present, or 1.11 grams of fluorine per pound of "Defluorite". This corresponds to a removal of 32.19 grams of fluorine per cubic foot, which is somewhat less than that of bone.

A number of other field studies were conducted throughout the state with activated alumina furnished also by the National Aluminate Corporation. This substance was also known as "Defluorite" at one time. The material was not found to function in the removal of fluorides to any marked degree. Results of these studies are shown in Figs. 21 and 22.

GENERAL DISCUSSION

The effectiveness of the use of bone in removing fluorine from water has been investigated. It has been found that bone will reduce the fluoride content but that the degree of removal is dependent on a number of factors, especially the fineness of subdivision of the bone and the time of contact of the bone and water. Probably the most important consideration is the preparation and activation of the bone itself. The experiments which have been conducted show that the best bone product from an all-around point of view is the 40 - 60 mesh bone prepared by the calcination process which has been described. This bone shows not only the greatest fluoride-removing power, but it has a number of other advantages as well, including less loss by attrition, greater ease of wetting, and no tendency to putrefy.

The two types of filters which have been developed have been designed to give the maximum amount of fluorine removal with the least possible effort on the part of the user. The "olla" filter has been designed mainly for the treatment of drinking water. This type of unit is simple in operation, low in expense, and at the same time very convenient for home and office use. The cartridges of bone can be easily and quickly changed whenever the capacity of the bone has been reached. The necessity for strict control, however, in the operation of these filters cannot be overemphasized. The amount of bone to be used in any case is a function of the fluoride content of the water, the rate of flow, and volume of water needed. For every new unit installed, the size of the filter should be correlated with the above mentioned conditions.

In the field work which was conducted at the Pima County Preventorium strict control was maintained by taking water samples each day and analyzing for the fluoride content. Whenever the effluent water reached a value of 0.9 part per million of fluorine, the toxic level, a fresh charge of bone was installed. Similar control should be exhibited over the filters used by the individual.

In the case of the operation of the olla filter, it is suggested that a small "test-kit" be included with the unit so that tests could be made by the user from time to time. This "test-kit" contains two 100 cc. Nessler tubes, a 5 cc. pipette, a bottle of acid-indicator mixture, and a large bottle of a 1 part per million fluorine solution. This is sufficient equipment to make a field test using the Sanchis method as modified by Scott* who has shown that the acids and the indicator solution may be combined and that boiling is not necessary for the production of the color. The test involves the following steps--(1) fill one Nessler tube with the effluent water and the other tube with the standard 1 part per million fluorine solution; (2) add to each tube 5 cc. of the acid-indicator** mixture by means of the pipette; (3) let the solutions stand for 1 hour and compare the colors obtained. A comparison of the effluent water with the standard shows the user the status of his bone. If the color of the water is pinker than the standard, the capacity of his bone has not been reached; however, if the color is yellower than the standard, the bone has been exhausted and a fresh charge or cartridge should be installed. A record kept of the number of gallons used indicates the time to change the bone in future cases.

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* Unpublished results.

** Dissolve 1 gram of zirconium nitrate in 100 cc. distilled water, and 0.2 gram of alizarin red S in 100 cc. distilled water, and mix the solutions. Prepare 1 liter of 2.6 N hydrochloric and sulfuric acids and mix. Transfer 70 cc. of zirconium-alizarin mixture to a 1 liter flask, and make up to 1000 cc. with the mixed acid solution. Use 5 cc. of this acid-indicator per determination.

It is estimated that these olla type filters including cooler and cartridge of bone could be placed on the market for about $15 - $20. Since it is known that the capacity of the bone will eventually be reached, a service must be maintained to supply fresh bone and to regenerate the spent bone.

the size of the cartridge used in the olla filter in the laboratory studies limited the amount of bone which could be incorporated to about one and one-half pounds. Larger cartridges and even larger coolers could be constructed such that the amount of water on reserve could be increased as well as the total volume of water treated per charge of bone.

The pressure type of filter has been designed for more rigorous use. This unit is best adapted to those conditions in which a large supply of water is needed in a short time. Here again, however, the amount of bone to be used is dependent on the initial fluoride content of the water, the rate of flow, and the volume of water needed. For general all-around domestic use including cooking as well as drinking, this type of filter is recommended. The size of the filter can be increased to fit the requirements placed upon it. A rather large filter, for example, would undoubtedly be required for use in a school where the consumption of water is, at intervals (play periods, etc.), rather rapid.

It has been shown that the length of life of the bone is increased by longer time of contact, which, in this case, would mean a slower flow rate. The capacity for fluoride removal of one of these types of filters could be increased by the construction and use of a reservoir tank. Water could be then passed through the filter at a reduced rate of flow into the tank thus building up a large supply of treated water, and, at the same time, increasing the length of life of the bone and the effectiveness of removal. A flow rate of 25-30 gallons or less per hour is recommended on a 10 pound unit because greater flow rates do not effect as good a removal, and the capacity of the bone becomes exhausted more quickly.

Since these units are placed directly in the water line, the necessity for control becomes greater than ever. For each new unit installed the amount of bone necessary should be correlated with the initial fluorine content of the water, the rate of flow desired, and the volume of water that is expected to be used per unit of time. Each filter should be equipped with a meter fitted with an automatic alarm to indicate that regeneration of the bone is necessary. The standard "test-kit" previously described should be supplied with these units. After the bone has become exhausted fresh charges should be installed.

A number of field and laboratory tests have shown the practicability

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Methods of removal:

It is evident that only two courses of events are open with which to cope with fluoride bearing waters -- change of the water supply to a non-toxic supply, or a removal of the fluorine in the water to a non-toxic level. For a municipal supply, if the change is feasible, the former method is, of course, the most practical, the simplest, and probably the least expensive. However, a change of supply in the case of the individual is often impossible or too expensive. Especially is this true of the arid sections where water is not always plentiful. It is imperative, then, that a method for removing fluorine from water be developed. This appears to be the only solution for the individual. The method developed should be simple and relatively inexpensive, if it is to be used on any widespread scale. Various methods for the removal of fluorine from water have been suggested since the discovery in 1931 that this element causes mottled enamel, but practically all have proven to be impractical for one reason or another.

Boruff in 1934 (9) included the use of aluminum sulphate, sodium aluminate, zeolite, activated alumina, and bauxite in a study on methods of removing fluorine from water. In concentrations between 2 to 3 parts per million of fluorine the application of 2 grams per gallon of sulphate of alumina reduced the fluoride content of the water to 1.0 per million. Sodium aluminate, zeolite, silica gel, sodium silicate, and ferric salts were not effective. He also noted that the addition of calcium hydroxide effected a substantial reduction of fluoride. Kempf, Greenwood, and Nelson (33) in studying the aluminum sulphate method emphasized the of the use of bone in the removal of fluorine from water. The cost of production of these units cannot as yet be determined. Steamed bone meal can be purchased for $2.75 per 100 pounds, but since the yield is only 50 - 60 per cent, the initial cost of the bone is nearly doubled. The added costs of chemicals, calcination, labor, and overhead must also enter, thus making any estimate merely conjectural.

The practicability of the use of bone on a municipal scale has not as yet been determined. But little reason is seen to prohibit its use, unless the cost of production and maintenance of such large quantities of bone should become too high. From a study of flow rates it can be shown that the size of filter bed necessary for the passage of 1,000,000 gallons of water/24 hours/12 inch depth of bone is 10 square feet in the case of 10-20 mesh bone, 60 square feet for 20-40 mesh, and 100 square feet for 40-60 mesh. Consider the application of the use of bone for fluorine removal in the case of a city using 1,000,000 gallons of water per day containing 3.5 parts per million of fluorine. It has been shown that 1 cubic foot of bone (40-60 mesh) will treat 4900 gallons of a water containing 3.5 parts per million of fluorine. The amount of bone necessary, therefore, to treat the 1,000,000 gallons of water is given by the ratio 1,000,000/4900 which is 208 cubic feet. The filter bed required for treatment of this water using 40-60 mesh bone would have to be at least 10 feet square and 2 feet deep, which is not beyond the realm of possibility considering that bone can be activated. If the depth of the filter bed is increased its area can be decreased, thus filtration by means of pressure may become necessary.

That phosphates are capable of removing fluorine from water has been shown by other investigators also. The work of Mac Intire and Hammond (37) and that of Adler, Klein, and Lindsay (1) indicates that several kinds of phosphates may function in this removal process. Other methods of removing fluorine from water have either proven ineffective or impractical for one reason or another with the possible exception of "Defluorite", a synthetic tri-calcium phosphate which has been put on the market by the National Aluminate Corporation of Chicago, Illinois, as a result of the work of Adler, Klein, and Lindsay cited above. Field tests conducted at the University of Arizona have shown that this material will function in the fluoride removal process but that its capacity is not so great as that claimed by the manufacturers. And it has further been shown on the basis of field studies that the fluoride removing power of bone per cubic foot is more than twice as great as that of "Defluorite". The bone method appears to be the most practical method for removing fluorides from water that has yet been devised.

SUMMARY AND CONCLUSIONS

1. The Sanchis method for the determination of fluorine in water was investigated and found to be affected by several ions. Phosphate ion causes low results and sulfate ion causes high results with this method.

2. Bone may be preprared for use in removing fluorine from water by boiling with alkali till the material has lost its flinty characteristics and has become snow-white in appearance, washing out the excess alkali with water, and neutralizing with a dilute hydrochloric acid treatment.

3. After use of the bone for the fluoride-removal process, it can be regenerated for further use by again treating with alkali and acid. An over-night treatment of the bone with cold alkali (0.25 N) followed by a ten-minute wash with a dilute acid such as acetic or hydrochloric acid (0.10 N) is recommended since less solubility loss occurs.

4. Calcination of the bone at 400 to 600 degrees centigrade for ten minutes followed by a ten-minute acid treatment yields a better bone product for the fluoride-removal process. Bone prepared in this manner will not putrefy as do some of the alkali-acid activated products.

5. The effect of mesh was investigated and was found to play an important part in the process of removal, the finer fractions removing the greater amount of fluorine. A 40 - 60 mesh has been found most desirable.

6. The time of contact of the bone and the water has been found to be of considerable importance, more fluorine being removed on the greater

time of contact.

7. The pH of the water has little effect on the amount of fluorine removed by bone over a range of pH 3 to 8. At higher pH values the removal is decreased, but since few domestic waters ever attain any such degree of alkalinity, the effect is considered insignificant.

8. Temperature has been found to have no effect on the fluoride-removing power of bone.

9. The effect of the presence of other salts than fluorine in waters on the amount of fluorine removed by bone was investigated, and was found to have only a very slight effect tending in some cases to decrease the amount of fluorine removed.

10. Double-treatment of water with two portions of bone was found to function no better than a single treatment with twice as much bone.

11. The mechanism by which bone removes fluorine from water appears to be through the formation of solid solutions rather than simple anion-exchange as was first supposed.

12. Fluorine-removal by other phosphates has been investigated, but none has functioned so well as bone. The synthetic tri-calcium phosphate product of the National Aluminate Corporation known as "Defluorite" was found to possess the capacity for fluorine removal, but on the volume basis bone has been shown to be more than twice as effective.

13. The practicability of the use of bone for removing fluorides from drinking water has been demonstrated by a number of field and laboratory studies.

14. Two types of filters for domestic use, the "olla" and "pressure" filters, have been presented and their practicability proven by a number

of field studies.

15. The bone method presented appears to be the most practical and economical method that has as yet been devised for removing fluorine from drinking water.

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necessity of exact pH control. This method was costly and necessitated the use of a skilled chemist, and even then results were often unreliable.

McKee and Johnson in 1934 (39) (patent No. 2,072,376) suggested the use of activated carbon. However, the method was considered to be impractical because of the high cost of the material and also because satisfactory removal could be accomplished only at a pH of 3. or under.

Fink and Lindsay in 1936 (24) used activated alumina (Churchill patent No. 2,059,553), commercially known as defluorite, in the laboratory and reported effective fluorine removal from water. However, this material did not prove effective in the field in Arizona. Latest reports from Dr. Fink indicate that they have abandoned the use of the material as being impractical.

Scott in 1937 (50) noted a lowering of the fluorine concentration in water by lime softening plants. He, therefore, suggested the use of lime in reducing the fluoride content of waters. The method, though, was found to have several serious limitations: (1) the fluorine removal was found to be a function of the magnesium removed, and, therefore, a salt of magnesium had to be added if not originally present in the water in sufficient quantity; (2) to effect a high percentage removal of magnesium, essential to secure proper fluoride reduction, necessitated adding sufficient lime to raise the pH to around 10.5; and, under these conditions, a caustic alkalinity of about 30 parts per million is required which must be corrected for by carbonation; and (3) the initial fluorine content of the water could not be over 3.3 parts per million if the method was to be used on a practical scale. For a village of about 1,000 population, Scott estimates a cost of about $10,000 for the construction of a water-softening fluoride-removing plant. It is obvious that this method would carry so much expense as to make it impractical.

More recently, 1938, the work of Mac Intire and Hammond (37) and of Adler, Klein, and Lindsay (1) shows that freshly prepared calcium phosphates of various kinds were capable of removing fluorine from water. Mac Intire and Hammond reported that the precipitation of solute fluorides as fluorapatite, $3Ca_3(PO_4)_2 \cdot CaF_2$, from suspensions of active forms of tricalcium phosphate can be utilized to effect complete removal of fluorides from ground waters. Either extended cold agitations, boiling, or filtration through sand-phosphate mixtures induces the fluoride removal. The most feasible procedure for small-scale operations is to introduce about 4 parts of either baking powder or superphosphate, preferably the "triple" type, into 1000 parts of water; the dissolved phosphates are then precipitated out of the boiled solution along with the fluorides by addition of a small excess of calcium hydroxide, which in turn is precipitated by aeration. Either prolonged settling or immediate filtration removes the phosphate-fluoride precipitate, it is claimed. Negative results, however, were obtained at Arizona using phosphoric acid and lime as suggested by Mac Intire (patent No.2,126,793).

Adler, Klein, and Lindsay (1) reported that their observations of the gel structure of tricalcium phosphate during the process of manufacture suggested the possibility of its use as an adsorbent for fluorides. Test towers containing small amounts of the freshly prepared, granular, tricalcium phosphate of 20 to 40 mesh size shows the material to have approximately twice the efficiency of activated alumina reported by Fink and Lindsay and investigated by Swope and Hess (64). Field tests making use of small gravity units carried out by other observers verify the results obtained by these investigators, it is claimed. One per cent sodium hydroxide solution followed by a dilute hydrochloric acid wash has been found to be effective for regeneration.

Clark and Mann in 1938 (14) proposed a method of fluoride removal from water using bone ash. They have taken commercial bone ash, which can be purchased already for use, and have heated water to which small amounts of this material was added. After heating, the bone ash settles to the bottom of the container. They report that about 80 per cent of the water samples so treated (waters not too high in fluorine) have had their fluorine content lowered from toxic to non-toxic quantities. For waters containing over 3.0 parts per million of fluorine a slightly different procedure was used. In this case the bone ash is added to the water and dissolved in hydrochloric acid. The solution is then neutralized with baking soda and the phosphate thus reprecipitated in flocculent form. This precipitate was filtered, washed, and used just as was the untreated bone ash mentioned above. They report that in every case tried this treated bone ash has effected an almost complete removal of all fluorine.

In 1934 H. V. Smith and M. C. Smith presented preliminary evidence that phosphates are capable of removing fluorine from water (55, 57). This possibility was suggested to these two investigators by W. T. McGeorge of the Arizona Agricultural Experiment Station. In 1937 further evidence was presented (58) in a preliminary study of the use of specially treated animal bone. The method proposed depends upon the affinity of phosphates for fluorine. This fact was first noted in 1893

FIGURES

Fig. 1. Typical Cases of Mottled Enamel Found in Certain Areas in Arizona.

Fig. 2. Distribution of Mottled Enamel in the United States.

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Fig. 20. "Pressure" Type Filter.

Fig. 21. Removal of Fluorine from Water Using Activated Alumina.

Fig. 22. Removal of Fluorine from Water at St. David School Using Activated Alumina.

THE USE OF BONE AND OTHER PHOSPHATES FOR THE REMOVAL OF FLUORINE FROM DRINKING WATER

by

William B. Davey

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